THE OLFACTORY SENSE OF COLEOPTERA

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CONTENTS.

INTRODUCTION AND METHODS.

In the investigation here recorded two objects have been kept in view: (1) To make a careful study of the morphology and physiology of the olfactory pores of beetles, and (2) to determine experimentally whether or not the olfactory organs lie in the antennæ.

Since those investigators, who have performed experiments on beetles with mutilated antennæ, have failed to study sufficiently the behavior of the insects investigated, the responses observed have misled them in determining the seat of the olfactory organs. Entomologists are generally agreed that the organs of smell in beetles lie in the antennæ, but when the results of those who have performed experiments on beetles are carefully considered, it is seen that some beetles with amputated antennæ smell practically as well as unmutilated ones, while other beetles are materially affected when the antennæ are mutilated. Hicks (1857 and 1860) discovered some peculiar organs (called olfactory pores by the present writer) on the wings and legs of beetles and he suggested that they have an olfactory function. Lehr[1] discovered the same organs on the peduncles of the elytra of *Dytiscus marginalis*. The present writer ('14*a* and *b*) made a comprehensive study of the olfactory pores in Hymenoptera and he ('14*c*) gives a complete review of the literature pertaining to the sense of smell in insects. The present paper embodies the results of a careful study of the olfactory pores in Coleoptera in much the same manner as pursued on those in Hymenoptera.

To obtain material for the study of the disposition of the olfactory pores, adult specimens were used. In regard to prepar-

[1] Lehr's paper, which deals only with the morphology of these organs, was overlooked until after my paper had been sent to press. Lehr has not seen any of my papers on this subject because my first one ('14*a*) appeared only three months before his, and my second one ('14*b*) appeared in the same month as his.

ing the specimens with caustic potash and to bleaching them with chlorine gas, the reader is referred to the writer's work on Hymenoptera ('14*b*, p. 295).

To obtain material for the study of the internal anatomy of the organs herein discussed, beetles just emerging from the last pupal stage were mostly used. At this stage the chitin is soft, the wings are usually expanded, and the sense organs are fully developed. In order that the desired stages of beetles might be had, many larvæ and pupæ of various Coleoptera were collected on plants and in rotten stumps and logs. These immature insects were reared in the laboratory. When each one of them had reached the proper stage, it was killed and parts of it were put into a fixing fluid.

The writer ('14*a*, p. 268) describes the usual method of embedding with celloidin and paraffin. Since then, a rapid method has been used which is described in detail as follows: The various appendages of the insects are removed, and are cut into small pieces, which are immediately dropped into a modification of Carnoy's fixing fluid. This fluid, containing equal parts of absolute alcohol, chloroform, and glacial acetic acid, with corrosive sublimate to excess, should be kept in a glass-stoppered bottle so that it may not lose its fixing ability by air being mixed with it. Also, while dropping material into vials containing this fluid, the stoppers of the vials should not be removed longer than absolutely necessary. When the material sinks to the bottom of the vial, it is removed and is thoroughly washed in 85 per cent. alcohol. It is then preserved in 85 per cent. alcohol. When ready for embedding, the material is cut into pieces from two to four millimeters in length. These pieces are then put into 95 per cent. alcohol containing eosin. When sufficiently stained, they are placed in a vial containing absolute alcohol and cedar oil. As soon as they sink through the alcohol into the oil and lie on the bottom of the vial, the alcohol and oil are removed. A small amount of ether is then poured into the vial. Five minutes later the ether is removed, and thin celloidin is poured into the vial. Ten minutes still later the thin celloidin is exchanged for thick celloidin. After remaining in the thick celloidin five minutes, the pieces of material are removed and are put into a vial of

chloroform where they remain five minutes. They are then embedded in 55° M.P. paraffin for five minutes. The sections were cut from five to ten microns in thickness and when they failed to ribbon the microtome knife was warmed. From this stage on the sections are treated like ordinary paraffin sections with the following exceptions. A rather thick film of fresh Mayer's albumen is spread upon each slide. After drawing the water from the slide upon which are mounted the sections, the latter are flattened to the slide by using a piece of wet tissue paper. No heat is used for straightening the ribbons on the slides because the least amount of heat blisters the celloidin. After drying over night, most of the sections adhere to the slides while being passed through the reagents, but to be sure of not losing any sections, the slides were sometimes wrapped in tissue paper and thread was then firmly wound around the paper. Instead of using absolute alcohol a mixture of equal parts of absolute alcohol and chloroform is employed so that the celloidin may not be dissolved, and instead of using eosin in 95 per cent. alcohol as a counter stain, the eosin is put into a mixture of the absolute alcohol and chloroform. The sections were stained in Ehrlich's hematoxylin from 10 to 15 minutes, the time depending on their thickness and whether or not they were wrapped in tissue paper.

The writer is grateful to Mr. H. S. Barber of the Bureau of Entomology for most of the dried specimens used which belonged to the collections of the U. S. National Museum. Mr. Barber is also to be thanked for the identification of all the beetles used in the experimental part of this work.

MORPHOLOGY OF THE OLFACTORY PORES.

Before experimenting to determine the function of the organs called the olfactory pores by the writer ('14*a*), the distribution and number of these pores in many beetles were studied.

DISPOSITION.

In making a comparative study of the disposition of the olfactory pores in beetles, 50 species, belonging to 47 genera and representing 34 families, were used. With the exception of two

species used for individual and sexual variations, only one specimen of each species was studied. Whenever a portion of an appendage or an entire appendage was missing or was badly mutilated in being prepared for study, the number of pores on this portion or entire appendage was regarded the same as the number found on the corresponding portion or entire appendage on the opposite side of the body. Since the pores on only one specimen for each species were counted, the total number of pores recorded can not be a fair average. Besides this error, there is also a probable error of not less than 10 per cent. on an average for all the specimens. In the smaller specimens the probable error is perhaps not more than two or three per cent., but in some of the larger ones, this error is probably more than 10 per cent. The pores on only the legs, elytra and wings are included in the total numbers. Other parts of the insects were not examined, and it is quite possible that olfactory pores may be found on some of the parts not examined, particularly on the mouth parts.

(*a*) *Epilachna borealis*.

Since the lady beetle, *Epilachna borealis*, is most conveniently studied and as its pores are typical for most of the smaller beetles, the disposition of its pores will be described in detail, and then the variations found in the other species will be given.

The elytra and wings have dorsal and ventral surfaces, and the legs may be divided for description into two surfaces. The inner surface faces the body of the beetle and the outer surface is directed from the body. On the specimen examined, one group of pores was found on the peduncle of each elytron; three groups besides a few scattered pores on each wing; and two groups besides a few scattered pores on each leg. The groups and scattered pores are located as follows: Group No. 1 lies on the dorsal surface of the peduncle of the elytron with its distal or broader end against or just beneath the basal margin of the elytron (Text-fig. 1*A*, *BM*). Under a high-power lens, it is seen that this group lies on the radial plate (Text-fig. 1*B*, *RP*) between the muscle disk (*MD*) and the subcostal head (*ScH*). The distal ends of these heavy chitinous plates sometimes lie beneath the

basal margin (*BM*) of the elytron so that all or a portion of the group may be concealed. In such a case it is necessary to pull the peduncle from beneath the base of the elytron in order to count the pores. Group No. 1 on the left elytron consists of 71 pores (Plate I., Fig. 4), while the same group on the right elytron has 78 pores.

Groups Nos. 2, 3 and 4 lie on the dorsal surface of the wing on the radius (Text-fig. 1*C*, *R*). No. 2 lies on the extreme anterior

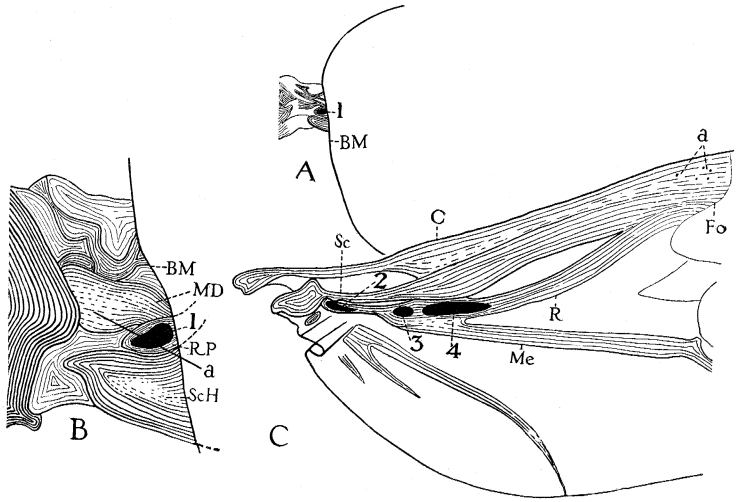

FIG. 1. Portion of left elytron and left wing of the lady beetle, *Epilachna borealis*, showing groups 1 to 4 of olfactory pores, as indicated by the numbers 1 to 4; *A* shows relative sizes of peduncle of elytron and group of pores on peduncle when compared with size of basal margin (*BM*) of elytron; *A* and *B*, dorsal surface of peduncle of elytron, showing position of group 1 of olfactory pores on radial plate (*RP*) between muscle disk (*MD*) and subcostal head (*ScH*). The lower side of each drawing is the outer margin of the elytron. *A*, × 8; *B*, × 45; *C*, dorsal surface of wing, showing position of groups 2 to 4 of olfactory pores on radius (*R*), × 8; *a*, position of scattered pores on ventral side of wing on union of costa (*C*) and subcosta (*Sc*) near fold of wing (*Fo*). Sometimes a group is found on the media (*Me*) just below group 4.

end of the radius and it is usually difficult to count its pores, because the surface of the radius at this place is greatly arched causing some of the pores to lie on the top of the arch while the remainder of them lie on the side of the arch facing the anterior margin of the wing. Nos. 3 and 4 are found on the radius where the media (*Me*) joins the radius. On the right wing, No. 2

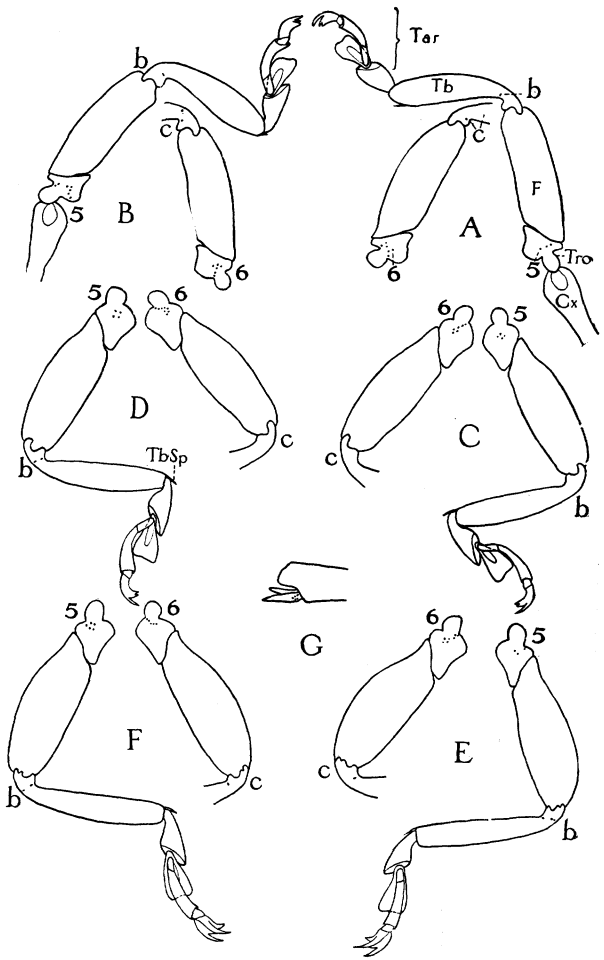

FIG. 2. Position of olfactory pores on legs of beetles, × 8; *A–F*, legs of lady beetle, *Epilachna borealis*, showing position of groups 5, 6, *b* and *c* of olfactory pores. The drawing of each leg in which the tarsus (*Tar*) is shown represents the outer surface of that leg, and the drawing not showing the tarsus represents the inner surface of the same leg. *A*, right front leg; *B*, left front leg; *C*, right middle leg; *D*, left middle leg; *E*, right hind leg; *F*, left hind leg. *G*, distal end of tibia from front leg of *Epicauta marginata*, showing five olfactory pores on one of the two tibial spines.

consists of 55 pores; No. 3 of 43 pores; and No. 4 of 43 pores. On the left wing, No. 2 consists of 50 pores; No. 3 of 43 pores; and No. 4 of 46 pores.

Group *a* of the scattered pores lies on the ventral surface of the

wing near the anterior margin of the wing a short distance from the place where the wing folds (Text-fig. 1*C*, *a*). On the right wing it consists of three pores and on the left wing of five pores.

Groups Nos. 5 and 6 are located at the proximal end of the trochanter (Text-fig. 2*A–F*, *Tro*), No. 5 lying on the outer surface and No. 6 on the inner surface. No. 5 usually extends only about half way across the leg, while No. 6 extends nearly all the distance across the leg. No. 5 on each leg consists of five pores, except on the left front leg where there are seven in it. On the right side No. 6 on each leg consists of seven pores, whereas on the left side on each of the middle and hind legs it consists of eight pores but of only six on the front leg.

Groups *b* and *c* of the scattered pores lie at the proximal end of the tibia (Text-fig. 2*A–F*, *Tb*), group *b* being located on the outer surface and group *c* on the inner surface. Group *b* on each front leg consists of only one pore; on the right middle leg it has one pore, but on the left middle leg it has two pores; on the right hind leg it has three pores while on the left hind leg it consists of two pores. Group *c* on each front leg has three pores, whereas on each of the middle and hind legs it has only one pore.

All six legs of the specimen of *Epilachna borealis* examined bear 95 olfactory pores; both elytra carry 149 pores, and both wings carry 288 pores. All of these combined make 532 olfactory pores.

(*b*) *Other Species.*

The greatest variation found in the olfactory pores of the other species examined is in regard to the total numbers of the pores. The second greatest variation is in regard to the distribution of the pores on the wings. This variation and other minor ones will now be given and a discussion of the total numbers of the pores will be presented last. For sake of brevity, instead of using the long scientific names of the beetles, the species will be numbered from 1 to 50, and those interested in associating the names of the species with the variations described may do so by referring to the names and numbers of the species in the table on page 419.

A group of pores (No. 1) was found on the peduncle of each elytron. This group in 46 species is definite, that is, the pores

are close together and are not scattered as they are in the other four species (Nos. 2, 34, 35, 44). In some beetles it is almost impossible to identify the various chitinous plates in the peduncles of the elytra, but as far as can be ascertained the definite groups of pores are located on the radial plates, while the scattered groups may spread over two or more of the plates. In shape these groups are round, oblong and triangular. The triangular-shaped ones are most common. As a rule, the more pores in this group, the smaller they are and the closer they are together. In three species (Nos. 23, 29, 36) the pores in this group are comparatively large, while those in the lady beetles are medium in size. *Osmus* with 12 pores on both elytra has the least number and *Hydrophilus* with 310 pores on both elytra has the largest number. In regard to the total numbers of pores on the elytra for the 50 species, the reader is referred to the table on page 419.

The three beetles, *Osmus*, *Clinidium* and *Cysteodemus*, are wingless. No rudiments of the wings were even found. The number of groups of pores on each wing of the other species varies from 1 to 4. Ten species (Nos. 1, 10, 13, 14, 16, 17, 18, 21, 22, 47) have only one group on each wing. One wing of *Lucidota* has one group while the other wing has two groups. Twenty-one species (Nos. 4, 5, 8, 11, 20, 24, 25, 26, 28, 30, 31, 32, 36, 38, 40, 42, 43, 45, 46, 49, 50) have two groups on each wing. Twelve species (Nos. 3, 6, 7, 9, 15, 23, 29, 33, 37, 39, 41, 48) have three groups on each wing. Three species (Nos. 12, 34, 35) have four groups on each wing. When only one group is present on each wing it usually occupies the position of Nos. 3 and 4 of *Epilachna borealis* on the radius (Text-fig. 1*C*). It may be no longer than No. 4 of *Epilachna*, or it may extend nearly all the distance to the fold of the wing (*Fo*). When two groups are present on each wing, one is similar to No. 2 of *Epilachna* and the other is similar to Nos. 3 and 4 united. The latter group may or may not extend all the way to the fold of the wing. In *Collops*, 20 pores were found on the ventral side of one wing besides the two groups on the dorsal surface. When three groups are found on each wing, they may be located like those of *Epilachna*, or two of them may lie on the radius and the third

one on the media. The largest one is similar to No. 4 of *Epilachna* and it may or may not extend all the way to the fold of the wing. When the third group lies on the media, as in *Orthosoma* (Plate II., Fig. 31), it occupies a position just beneath the larger group on the radius. Its pores are generally scattered considerably. When four groups are found on each wing, one of them lies on the subcosta, two on the radius and one on the media. It is common for the distal end of the largest group on the radius of any wing to become attenuated so that a row of pores may extend nearly, if not all, the way to the fold of the wing. The farther this row of pores extends along the radius the farther apart are the pores. It is also common for the largest group on the radius to consist of pores of two sizes. The diameters of the larger pores may be two or three times those of the smaller ones. The larger pores extend lengthwise through the center of the group. Eight species (Nos. 1, 3, 4, 5, 6, 33, 36, 38) have pores as just described. The pores in this group of seven other species (Nos. 10 to 14, 23, 30) are also of two sizes, but there is not such a great difference in the sizes of the smaller and larger pores, as in the pores of the preceding eight species. These pores are also comparatively larger. All the pores on the wings of nine species (Nos. 7, 9, 16, 17, 18, 20, 22, 31, 40) are of about the same size and they are comparatively large. All the pores on the wings of the remaining species are of about the same size, but they are comparatively small. *Coxelus*, the smallest beetle examined, with 130 pores on both wings has the least number, while *Orthosoma*, perhaps the largest beetle examined, with 982 pores on both wings has the greatest number.

The trochanters never fail to possess at least a few pores. The trochanter with the fewest pores has two, whereas the one with the most has 59. As a rule, the more pores on a trochanter, the smaller they are. The pores are generally located at the proximal end of this segment in about the same arrangement as represented in *Epilachna* (Text-fig. 2*A*–*F*), but occasionally they are considerably scattered, and a few may be found at the distal end of the segment.

A pore was found at the proximal end of one or more femurs belonging to each of 18 species (Nos. 1, 2, 4, 7, 8, 10, 11, 13, 16,

17, 18, 20, 21, 22, 24, 31, 48, 49), and from one to three pores were found at the proximal end of each femur of *Elater*.

While it is common to find one or more pores at the proximal end of a tibia, many of these segments are entirely devoid of olfactory pores. The greatest number of pores found on any tibia at this place is nine. In each of the tibio-tarsal articulations of the front and middle legs belonging to *Cotinis* from 7 to 11 pores were found. Pores were found in the tibial spines (Text-fig. 2*G* and Plate II., Fig. 27) of 15 species (Nos. 9 to 11, 20 to 25, 31, 32, 34, 35, 45, 48). The pores usually lie on the bases of the large spines. The largest number of pores found on a single tibial spine is 12. Of the 50 species examined, *Passalus* has the most pores on these spines.

Pores were found on the tarsi of 13 species (Nos. 1, 2, 4, 10 to 12, 16, 18 to 21, 25, 31). The greatest number found on a single tarsus was 37. *Osmus*, one of the three apterous species, has the most pores on its tarsi.

Eleven species (Nos. 6, 26, 27, 29, 31, 32, 33, 38, 42, 44, 47) were found with no pores on the legs except those on the trochanters and on the tibial spines. *Cybister* with 49 pores on all six legs has the least number on these appendages, while *Podabrus* with 341 pores on all six legs has the largest number.

No special examination was made to find any structure other than the olfactory pores, nevertheless, minute pores were seen in 15 species (Nos. 7, 8, 14, 15, 16, 20, 25, 27, 29, 36, 40, 44, 45, 46, 48). These pores were seen on various parts of the beetles, but particularly on the legs and elytra. They usually lie near the bases of the hairs, but sometimes they lie a considerable distance from the hairs. Since they are many times smaller than the olfactory pores, without exception they are probably the pores belonging to hypodermal glands, as will be shown for those of *Epilachna* on page 423. However, a careful comparative study of these pores is needed before anything definite can be said about them.

Coxelus, the smallest species examined, has a total number of 273 pores which is the smallest number of all the winged species, while *Orthosoma*, perhaps the largest species examined, has a total number of 1,268 pores, which is the largest number of all the

species examined. As a rule the smaller the species, the larger are the pores, comparatively speaking, and the fewer they are. Likewise, the larger the species, the greater is the number of its pores and the smaller they are. As a rule there are no generic and specific differences, except variations in number of pores, the amount of variation depending on the size of the individuals compared. Judging from the sizes of the four water beetles examined, the pores on their legs are fewer and smaller than those on the legs of any other beetle examined. Pores were found only on the trochanters of *Cybister*, while a few were also seen on the femurs and tibiæ of the other three water beetles. The number of pores on the legs of these beetles are as follows: *Cybister*—49, *Dineutes discolor*—65, *Dineutes vittatus*—98, and *Hydrophilus*—93. These numbers indicate that the better the legs are adapted for locomotion in water, the fewer pores they have.

The small total numbers of pores of *Osmus*, *Clinidium* and *Cysteodemus* are due to the absence of wings. In *Osmus* and *Clinidium* more pores are found on the legs than might be suspected. The tarsi of *Osmus* have more than the tarsi of any other beetle while the tarsi of *Clinidium* have more than suspected.

The following table (p. 419) includes the family, name and number, the olfactory pores on the legs, elytra, wings, and the total number of pores of each of the 50 species examined. In the preceding pages the beetles are usually referred to in this table by their respective numbers.

(*c*) *Individual and Sexual Variations.*

For this study five males and five females each of *Harpalus pennsylvanica* and *Leptinotarsa 10-lineata* were used. No individual and sexual variations were found, except slight variations in the number of pores. The total numbers of pores of the males of *Harpalus* vary from 550 to 580 with 570 as an average; those of the females of *Harpalus* from 575 to 699 with 628 as an average. The average number of pores for males and females of *Harpalus* is 599. The total numbers of pores of males of *Leptinotarsa* vary from 665 to 780 with 722 as an average; those of the females

TABLE I.

THE NUMBER OF OLFACTORY PORES ON THE LEGS, ELYTRA AND WINGS OF COLEOPTERA.

Family.	Name and Number of Species.	No. of Pores on Legs.	No. of Pores on Elytra.	No. of Pores on Wings.	Total No. of Pores.
Cicindelidæ.....	1. *Cicindela vulgaris*	180	29	924	1,133
	2. *Osmus* sp.	290	12		302
Carabidæ.....	3. *Calosoma scrutator*	140	62	869	1,071
	4. *Harpalus caliginosus*	180	69	600	849
	5. *Harpalus pennsylvanica*	107	39	453	599
Dytiscidæ.....	6. *Cybister fimbriolatus*	49	180	917	1,046
Gyrinidæ.....	7. *Dineutes discolor*	65	46	435	546
	8. *Dineutes vittatus*	98	42	532	672
Hydrophilidæ....	9. *Hydrophilus triangularis*	93	310	662	1,065
Silphidæ.....	10. *Necrophorus marginatus*	111	60	652	823
	11. *Silpha inæqualis*	118	30	664	812
Staphylinidæ ...	12. *Staphylinus macrulosus*	110	23	493	626
Scaphidiidæ.....	13. *Scaphidium quadguttatum*	111	96	185	392
Coccinellidæ.....	14. *Coccinella 9-notata*	93	132	310	535
	15. *Epilachna borealis*	95	149	288	532
Endomychidæ...	16. *Endomychus biguttatus*	144	31	178	353
Erotylidæ.....	17. *Megalodacne heros*	102	120	383	605
Colydiidæ.....	18. *Coxelus guttulatus*	93	50	130	273
Rhyssodidæ.....	19. *Clinidium sculptile*	131	40		171
Cucujidæ.....	20. *Cucujus clavipes*	107	104	325	536
Mycetophagidæ .	21. *Mycetophagus punctatus*	165	135	379	679
Dermestidæ.....	22. *Dermestes marmorata*	90	80	570	740
Histeridæ.....	23. *Hister depurator*	74	80	249	403
Trogositidæ.....	24. *Tenebroides castenea*	96	113	296	505
Elateridæ.....	25. *Elater apicatus*	111	130	365	606
Buprestidæ	26. *Melanophila longipes*	186	110	278	574
Lampyridæ.....	27. *Lucidota californica*	177	123	257	557
Telephoridæ.....	28. *Chaulcognathus pennsylvanica*	308	157	445	910
	29. *Podabrus comes*	341	157	280	778
Malachidæ.....	30. *Collops bipunctatus*	120	101	248	469
Lucanidæ.....	31. *Platycerus quercus*	158	160	536	854
	32. *Passalus cornutus*	203	184	782	1,169
Scarabæidæ.....	33. *Canthon lævis*	62	180	724	966
	34. *Cotinis nitida*	162	39	934	1,135
	35. *Euphoria sepulchralis*	90	36	613	739
	36. *Osmoderma scabra*	127	182	782	1,091
Cerambycidæ....	37. *Orthosoma brunneum*	79	207	982	1,268
	38. *Callimoxys fuscipennis*	87	175	510	772
	39. *Cyllene robiniæ*	110	40	629	779
Chrysomelidæ...	40. *Leptinotarsa 10-lineata*	115	130	476	721
Bruchidæ.....	41. *Bruchus pisi*	65	137	352	554
Tenebrionidæ....	42. *Tenebrio molitor*	66	145	462	673
	43. *Uloma impressa*	80	119	361	560
Meloidæ.....	44. *Cysteodemus armatus*	133	39		172
	45. *Epicauta marginata*	157	94	504	755
	46. *Epicauta pennsylvanica*	125	100	440	665
Rhipiphoridæ....	47. *Myodites scaber*	154	260	350	764
Rhynchophora:					
Rhynchitidæ. .	48. *Rhynchites bicolor*	84	148	480	712
Otiorhynchidæ	49. *Cephus latus*	71	111	382	564
Curculionidæ .	50. *Zygops seminivius*	65	116	392	573
Variation		49–341	12–310	130–982	273–1,268[1]

[1] The total number of pores of apterous species are not included.

of the same species from 661 to 785 with 720 as an average. It is thus seen that the females of *Harpalus* have a few more pores than the males, while the males and females of *Leptinotarsa* have the same number of pores.

STRUCTURE.

In the preceding pages it has been shown that most of the variations in regard to the disposition of the olfactory pores are slight. In the following pages it will be shown whether or not this is true for the structure of these pores.

(*a*) *External Structure.*

When examined under a low-power lens, the olfactory pores may be easily mistaken for hair sockets from which the hairs have been removed. When more carefully observed under a high-power lens, a striking difference in external form is usually seen, but sometimes it is difficult to distinguish the pores from hair sockets. The pores appear as small bright spots when a strong transmitted light is used. Each bright spot has a dark boundary or pore wall (Plate I., Fig. 1, *PorW*).[1] Near the center of this boundary is a transparent spot, the pore aperture, which may be round, oblong, slit-shaped, or club-shaped. On the legs the pore apertures may be round (Fig. 2, *PorAp*), oblong (Fig. 3, *PorAp*), slit-shaped or club-shaped (Fig. 1, *PorAp*). On the elytra and wings they may be round or oblong (Figs. 4 to 8). The hair sockets (Figs. 1 and 2, *PorWHr*) are generally smaller than the olfactory pores and the pores of the hypodermal glands (Figs. 1 and 2, *PorWGl*) are easily distinguished from the hair sockets and olfactory pores by their small size.

(*b*) *Internal Structure.*

All the olfactory pores studied are more or less flask-shaped structures. They are of three general types. In the most common type, as found in *Uloma*, the mouth of the pore (Figs. 9–12, *Mo*) is flaring and the sense cell (Fig. 12, *CS*) lies in the lumen of the appendage outside the pore cavity. The chitinous

[1] All figures, except Text-figs. 1, 2, and 3 are numbered consecutively on Plates I. and II.

cone (Fig. 9, *Con*) never occupies more than one fourth of the pore cavity and usually much less (Fig. 12, *Con*). The cone always stains less deeply than the surrounding chitin, and it is common to see a hypodermal secretion (Figs. 9 and 10, *HypS*) inside the pore cavity. The sense fiber (Fig. 9, *SF*) pierces the cone, and the chitin between the pore aperture and the cone, and it ends in the bottom of the pore aperture or pit (Figs. 9–12, *P*) with its peripheral end exposed to the air in the pit.

The second type of pores is found in the legs of *Orthosoma* (Figs. 13–15), although the pores in the elytra (Fig. 21) and wings (Fig. 31) of the same beetle belong to the first or most common type. The chitinous integument of the legs of *Orthosoma* is thicker than that of the legs of any other beetle examined. Instead of the sense cells (Fig. 13, *SC*) lying in the lumen of the legs outside the pore cavities, in this type they lie inside the pore cavities. When the chitin forming the wall of the pore is not thick enough to protect the entire sense cell, the wall of the pore projects flange-like (Fig. 14, *Fl*) into the lumen of the leg. In Fig. 14 only about one third of the sense cell (*SC*) is shown. Studies of the olfactory pores in various hymenopterous insects made by the writer have shown that the sense cells begin to differentiate at the time when the chitin is beginning to be formed. From this fact, it is quite probable that the sense cells found in the second type of pores have not migrated into the pore cavities, but they now remain in approximately the same position as when the chitin was being formed.

The third type of pores is found in the legs of the lady beetle, *Epilachna borealis*. Instead of the chitin over the external end of the pore being depressed to form a pit, it is elevated dome-like above the surface of the leg. In the center of the dome lies the pore aperture (Fig. 16, *PorAp*). All the pores in the trochanters and most of those in the tibiæ (Fig. 17, *PorAp*) are of this type. Sometimes in the tibia is found a pore whose aperture is on a level with the surface of the tibia. The apertures of all the pores in the elytra (Fig. 18, *PorAp*) and wings (Fig. 19) of this beetle are on a level with the surfaces of the appendages.

As already stated, the olfactory pores of beetles are more or less flasklikė as a rule, but there are many variations among

them. They may be inverted flask-shaped as found in the legs of *Epilachna* (Figs. 16 and 17) and in the wings of *Passalus* (Fig. 20). Some have the shape of a flask without the neck (Figs. 9, 10 and 12). Some are long and slender like fingers or test tubes (Figs. 11, 18, 19 and 21).

Their sizes also vary much. The length of a pore always depends on the thickness of the chitin. The diameters of the pores of a small beetle (Fig. 25) may be as large, or even larger (Figs. 9 and 10) than the diameters of the pores of a large beetle (Figs. 13–15).

A chitinous cone is always present, although it may sometimes be almost indiscernible. It invariably has the same shade of coloration (Fig. 17, *Con*) as the remaining chitin (Fig. 17, Ch_2) which is formed after the insect has emerged into the imago stage. This is the first time that the writer has been able to determine definitely the formation of the cones. In all the hymenopterous insects studied by the writer, the chitinous integument is practically developed when the insects emerge, but in most beetles only about one third of the chitin is formed when the insects emerge. Since this is true the hypodermal cells are still large and they are rapidly secreting a substance which forms new chitin. Their external ends stand in contact with the chitin, and when no chitin is present they send processes into all holes or cavities in the chitin. Thus the hypodermal cell (Fig. 23, *HypC*) at the mouth of each olfactory pore sends a process into the pore. Since the sense fiber has entered the pore aperture before the cone is formed, the latter is formed at the external end of the pore around the sense fiber. When the chitinous integument (Fig. 17) is fully developed no hypodermal processes run into the pores and the hypodermal cells are very small.

The sense cells are always spindle-shaped (Figs. 12, 13, 16–19 and 23, *SC*). Only occasionally is an entire sense cell seen in a cross section, because the entire cell seldom lies in the same plane as that of the section. More entire sense cells may be seen in longitudinal sections, but even in these the cells are usually cut in two. Entire sense cells were best seen in the oblique sections through the peduncles of the elytra of *Passalus* and *Epilachna*. The nucleus (Figs. 13 and 23, *SCNuc*) of the sense cell is always

conspicuous. It may be darker (Fig. 13, *SCNuc*) or lighter (Fig. 23, *SCNuc*) in color than the cytoplasm in the cell. The nucleoli (Fig. 23, *SCNuc*) are also conspicuous.

Smaller sense cells may be seen in the sections through the proximal ends of the trochanters and through the proximal ends of the tibiæ. These (Fig. 17, SC_1) belong to tactile hairs (Fig. 17, *THr*).

In the sections through the legs and elytra of *Epilachna*, gland cells (Fig. 17, *GlC*) are plainly seen in the hypodermis (*Hyp*). These are equally as large as the olfactory sense cells, but they are quite different in structure. The diameters of the pores of the glands (*PorGl*) are slightly smaller than those of the hairs (*PorHr*), and they are much smaller than those of the olfactory pores (*Por*). The morphology and physiology of these gland cells will be given in another paper.

The shapes of the external ends or tops of the pits depend on the shapes of the pore apertures when seen in superficial views. That is, they are round, oblong, slitlike or clublike. The internal ends or bottoms of the pits are always round. The pore aperture, proper, is the round opening leading from the bottom of the pit to the external end of the pore. This aperture is closed by the peripheral end of the sense fiber. The shapes of the pits in cross sections, therefore, depend on the directions in which the microtome knife passes through the pits. The most common shape of a pit in cross section is that of an urn (Fig. 9, *P*). Pits including the pore apertures may be likened to round funnels, or to funnels slightly flattened, or to funnels considerably flattened, or to funnels so flattened that their tops would be club-shaped. In spiders the pits are slits which pass entirely through the cuticula. The sense fibers enter the pore apertures at the bottoms of the slits. The pits or slits in spiders, therefore, may be likened to funnels considerably flattened. When just emerged into the imago stage the pits (Fig. 9, *P*) in the legs generally extend about one-third the distance through the chitin, but when the chitin is fully developed, the pits extend perhaps from one fifth to one eighth the distance through the chitin. In all the figures showing two shades in the chitin, the darker one (Fig. 17, Ch_1) represents the chitin formed at the time when the

insect emerges from the last pupal stage, and the lighter one (*Ch*$_2$) represents the chitin formed after emerging into the imago stage.

As already stated, instead of the olfactory pores of the lady beetle, *Epilachna*, having pits, the chitin over each pore in the legs is elevated domelike above the surface of the leg. The olfactory pores (Figs. 24 and 25) in the legs of the two blister beetles, *Epicauta marginata* and *Epicauta pennsylvanica*, have only indications of pits. Their pore apertures are therefore on a level with the surface of the legs. The olfactory pores in the legs of the potato beetle, *Leptinotarsa 10-lineata*, have shallow pits (Fig. 26, *P*). All four just enumerated species have hypodermal gland pores distributed over the entire body except the wings. These pores are perhaps most abundant on the elytra, but they were never seen on the peduncles of these appendages, and it is quite probable that the secretion from their glands never covers the olfactory pores found on the wings and on the peduncles of the elytra. Judging from the gland pores, the hypodermal glands in the legs of *Epilachna* are more highly developed than are those of the other three species. The gland pores (Figs. 1, 2 and 28, *PorWGl*) on the legs of *Epilachna* lie on all sides and even among the olfactory pores, but in the legs of the other three species the gland pores never lie near the olfactory pores. When examined under a low-power lens the legs and elytra of *Epilachna* appear wet, and many small yellow flakes may be seen on them. The wet appearance is certainly due to the secretion from the hypodermal glands and the flakes are the remains of the secretion after it becomes dry. Thus in *Epilachna* there seems to be a direct correlation between the olfactory pores and the gland pores. Since the pore apertures in the legs lie above the surface of these appendages, the secretion from the hypodermal glands runs away from the pore apertures instead of into them. Such a device enables both sets of organs to function normally without the one hindering the other.

In the legs the sense cells always lie in a blood sinus (Figs. 16 and 17, *BlSin*) some distance from the muscles (Fig. 28, *M*). The nerves (*N*) are easily seen and branches (*NB*) are given off which run to the sense cells (*SC*). The neurilemma (Fig. 17,

Neu) of the nerve is usually distinct. In the cross section of a nerve, the nervous substance appears more or less netlike and nuclei, probably neuroglia nuclei (Fig. 17, *NeurNuc*), stand out conspicuously in the network. The trachea (Figs. 16, 17 and 28, *Tr*) and nerves (*N* and *NB*) are firmly suspended by the connective tissue whose nuclei (*ConTNuc*) are seen only occasionally. The lumen of the leg at the proximal end of the tibia of *Epilachna* seems to be divided into two chambers by a membrane (Fig. 17, Hyp_1) which resembles hypodermis. This structure has never been seen before by the writer and nothing can be said about its function.

The hypodermis (Fig. 18, *Hyp*) beneath the olfactory pores in the peduncles of the elytra is much thicker than elsewhere. It usually contains all the sense cells (*SC*), but in the elytra of *Passalus* the hypodermis is thinner and since the sense cells are so large and so numerous there is not enough room for all of them in the hypodermis. For this reason only a few of them lie among the hypodermal cells and the remainder of them lie in the lumen of the peduncle between the hypodermis and nerve. As usual they are surrounded by blood. In only one instance was the writer able to trace a sense cell all the way from the pore aperture to the nerve. Fig. 23 represents this sense cell connecting with the pore aperture (*PorAp*) and with the nerve (*N*). The trachea (*Tr*) lies by the side of the nerve. A large nerve (Fig. 29, *N*) and a large trachea (*Tr*) run through the radial plate (*RP*) of the peduncles beneath the olfactory pores. From the nerve many branches are given off which connect with the sense cells.

The hypodermis (Fig. 22, *Hyp*) beneath the olfactory pores in the wings is usually much thicker than elsewhere, but it does not contain the sense cells (*SC*). These cells lie in a blood sinus (Fig. 22, *BlSin*) between the hypodermis (*Hyp*) and the trachea (*Tr*), nerve (*N*) and nerve branches (*NB*). In the wings it is usually difficult to trace a sense fiber all the way to the pore aperture, but in oblique superficial sections this is easily done (Fig. 30). A large nerve and a large trachea run into each wing. These divide so that a smaller nerve and a smaller trachea run through each main vein.. The largest trachea (Fig. 31, *Tr*) runs

through the subcosta (*Sc*) while the largest nerves (*N*) pass through the veins bearing the olfactory pores. The nerve and trachea run directly beneath the sense cells (*SC*) and from the nerve pass off many branches which connect with the sense cells. In the costa (*C*) and subcosta (*Sc*) where there are no sense cells, only a few nerve fibers can be seen.

In the preceding pages it has been shown that there are many variations in the structure of the olfactory pores of beetles, and that these organs are very similar to those of hymenopterous insects. On the basis of the location of the pore apertures in the integument, the olfactory organs in beetles are intermediate between those of spiders and those of Hymenoptera.

EXPERIMENTS TO DETERMINE THE LOCATION OF THE OLFACTORY ORGANS.

Since it is now generally believed that the olfactory organs of beetles are borne by their antennæ, these appendages of many individuals were pulled off. From one to seven days later, the mutilated insects were tested with odors. In the preceding pages it has been shown that the olfactory pores of Coleoptera are located on the peduncles of the elytra, on the wings and on the legs. In order to ascertain if these structures receive odor stimuli, the elytra, wings and legs were mutilated. One or more days later these mutilated beetles were tested with odors. In all the experiments with unmutilated and mutilated beetles, 434 individuals have been tested. These belonged to 11 species representing eight families.

In order that the behavior of the mutilated beetles would be correctly interpreted, the behavior of unmutilated beetles under experimental conditions was first studied. Since it was not desired to ascertain the relative sensitiveness of males and females, both sexes were used indiscriminately. To determine the relative sensitiveness of unmutilated and mutilated individuals under conditions which permitted of their close observation, triangular experimental cases were employed. These were made of three narrow wooden strips, two of which were five and the third four inches long, each strip being half an inch thick. Wire screen served as a bottom and glass as a top for the case.

The apices and bases of these cases rested on two supports above a rigid table near a window. No screen was used to prevent the beetles from seeing the observer because they never showed any responses to the movements made by the observer.

The following sources of odors were used for determining the reactions of the beetles in the experimental cases; chemically pure essential oils of peppermint, thyme, and wintergreen; parts of plants—leaves and stems of pennyroyal (*Hedoma pulegioides*?), and of spearmint (*Mentha spicata*); decayed matter —parts of decayed beetles (*Harpalus pennsylvanica*). All these substances were kept in stoppered vials of the same shape and size. The leaves and stems of the pennyroyal were dried, but they still gave off a strong odor when the vial was uncorked. The leaves and stems of the spearmint were fresh and they did not emit as strong an odor as did the other substances used Beetles were killed and were torn to pieces. The pieces were put into a vial and after two or three days they emitted a foul and sickly odor.

A beetle was carefully placed into one of the experimental cases. When first put into the case the insect usually wandered about for several minutes, but finally it became quiet. The insect was tested with the above odors only when it had become perfectly quiet, without the antennæ being moved in the least. The stopper of a vial was quickly removed and the vial was gently and slowly placed under the experimental case directly beneath and within one half inch of the individual being tested. When all of these precautions are taken, a normal beetle generally responds to anyone of these odors within 60 seconds, but when all the reaction times are counted, it is seen that several of them failed to respond within 60 seconds. If a beetle when tested fails to react to an odor within 60 seconds, the response may be regarded as negative, and when it reacts to an odor within 60 seconds, the response may be called positive. As a control, an empty and odorless vial was now and then placed under the insects in the same manner. If by chance a beetle moved while the control test was being made, its behavior was different from that observed when odors were used. Only the first responses have been recorded and in all cases where there was the least

doubt as to whether the insect moved for any reason other than the olfactory stimulus, such movements were never recorded. The reaction time was counted in seconds. With an ordinary watch the minimum time which can be definitely recorded is two seconds, although many of the individuals responded to some of the odors much more promptly. Owing to this source of error, the average recorded time is probably double what it should be in the cases where all the responses for the same insect were prompt. An intermission of 10 minutes elapsed between any two tests in the same experimental case. Each individual was tested only once with the same odor.

In recording the responses the term "vibrated" is used to describe the rapid movement of the antennæ or legs up and down or from side to side. When this movement is slow, these appendages are described simply as having "moved." When the antennæ, legs or mouth parts are moved so that they are quickly bent at their articulations, they may be described as being "worked." When at rest a beetle usually lies flat on its thorax and abdomen, so the word "arose" means that the insect gets up and stands on its feet. In the averages of reaction times the probable error is presumably high. It has not been calculated since slight differences in reaction times are not considered as significant in the discussion of results. All anthropomorphic terms are put in quotation marks.

CARABIDÆ.

The Olfactory Sense of *Harpalus pennsylvanica*.

Many ground beetles (*Harpalus pennsylvanica*) were caught under flat stones in a corn field near the laboratory. As soon as brought to the laboratory, 25 of them were placed singly into the experimental cases. As they were being placed into the cases, some of them discharged a substance, presumably from the anal glands, which gave off an odor similar to that from formic acid. Confined in these cases, they sought the dark corners of the cases and did not wander about much inside the cases unless irritated. When half hidden in the dark corners, they rarely responded to odors, so it was necessary to keep them out of the corners while they were being tseted. The longer they remained

in the light and the more they were handled, the more satisfactory they were to experiment with. Owing to this kind of behavior, this species and several others used responded more slowly to odors a short time after being caught than they did a few days after being kept in confinement. This fact will explain why some unmutilated beetles just caught respond to odors more slowly than they do two or three days later after having had their antennæ pulled off. The following are the responses of this ground beetle to the odors from the six different substances and the average reaction times in seconds.

Oil of peppermint:

5 moved away quickly.	1 worked legs.
5 vibrated antennæ.	1 kicked quickly.
4 arose quickly.	1 vibrated antennæ and legs.
4 moved slightly.	1 vibrated legs.
2 moved antennæ and legs.	1 jumped slightly.

Reaction time 2 to 10 seconds, average 3.6 seconds.

Oil of thyme:

6 moved away quickly.	1 arose slowly.
5 moved quickly.	1 vibrated antennæ.
5 moved slightly.	1 moved backward slowly.
2 worked antennæ.	1 worked legs.
2 moved antennæ and legs.	1 did not respond.

Reaction time 2 to 60 seconds, average 8.5 seconds.

Oil of wintergreen:

5 moved away quickly.	1 stroked antennæ.
5 moved slightly.	1 vibrated antennæ and legs.
4 moved away slowly.	1 worked antennæ.
3 moved quickly.	1 worked legs.
2 vibrated legs.	1 did not respond.
1 arose slowly.	

Reaction time 2 to 60 seconds, average 16.4 seconds.

Leaves and stems of pennyroyal:

10 moved away quickly.	1 moved away slowly.
6 moved slightly.	1 vibrated legs.
3 vibrated antennæ.	1 did not respond.
3 worked antennæ.	

Reaction time 3 to 60 seconds, average 21.1 seconds.

Leaves and stems of spearmint:

5 moved away slowly.	2 did not respond.
5 moved slightly.	1 worked mouth parts.
3 moved antennæ and legs.	1 vibrated legs.
2 moved away quickly.	1 vibrated antennæ and legs.

2 worked antennæ.
2 jumped slightly.
1 moved antennæ.

Reaction time 3 to 60 seconds, average 21.8 seconds.

Parts of decayed beetles:

7 moved slightly.
5 did not respond.
4 moved away quickly.
3 moved away slowly.
2 jumped slightly.
1 moved legs.
1 worked antennæ.
1 vibrated legs.
1 vibrated antennæ and worked mouth parts.

Reaction time 5 to 60 seconds, average 28.1 seconds.

The general average reaction time of the 25 beetles tested to the six odors is 16.5 seconds. As a possible reason why one fifth of the individuals tested failed to respond to the odor from the decayed beetles is that these insects probably do not respond to decayed matter unless they are hungry. The 25 beetles tested were put into a wooden box four inches wide, seven inches long and two inches deep. One half inch of moist earth was also put into the box. The beetles soon buried in the earth and from that time on they appeared quite "at home." The box was put into a table drawer where it was more or less dark. About twice each week water was poured upon the earth and the beetles were fed earthworms and various insect larvæ. They drank some of the water and always greedily ate the food given to them. Up to the time of this writing (Jan. 15), 24 of these beetles have died. These lived from 18 to 180 days with 61 days as an average. All the beetles confined in the laboratory have not been fed since Oct. 15, but they have been given water once or twice a week. A few of the dead beetles when removed from the box had been partially eaten, but these insects were never seen fighting one another. While collecting this species in the corn field, a dead one was now and then found.

(*a*) *Effects with Antennæ Pulled Off.*

The antennæ of 25 *Harpalus pennsylvanica* were pulled off at their bases. These insects were then put into a wooden box similar to the one containing the unmutilated individuals just described. This box, also containing moist earth, was placed into the table drawer. The beetles appeared normal in all respects for they drank and ate as greedily as the unmutilated

ones and buried in the earth as usual. Seven days later they were placed singly into the experimental cases and were tested with the six odors as usual. They wandered about in the cases slightly more than did the unmutilated ones, but when tested they gave similar responses and reacted just as promptly.

Their reaction times are as follows: Oil of peppermint, 2 to 15 seconds, average 3.8 seconds; oil of thyme, 2 to 25 seconds, average 4.7 seconds; oil of wintergreen, 2 to 25 seconds, average 6.9 seconds; leaves and stems of pennyroyal, 3 to 50 seconds, average 14.4 seconds; leaves and stems of spearmint, 3 to 60 seconds, average 34.9 seconds. Ten failed to respond to this odor. Parts of decayed beetles, 3 to 60 seconds, average 32 seconds. Eight failed to respond to this odor. The general average reaction time of the 25 beetles tested to the six odors is 16.1 seconds. Up to the time of this writing (Jan. 15), 23 of these beetles have died. They lived from 19 to 171 days with 58 days as an average.

(*b*) *Effects with Elytra and Wings Pulled Off.*

The elytra and wings of 25 *Harpalus pennsylvanica*, just collected from the cornfield, were pulled off at their articulations. These mutilated insects were then put into a third box, similar to the two already described. The box was kept in the table drawer with the others. On the following day after mutilating the beetles, they were placed singly into the experimental cases and were tested with the six odors as usual. They seemed normal in all respects except they were extremely restless. Their responses to odors were similar to those of unmutilated ones, except they were slower.

Their reaction times are as follows: Oil of peppermint, 3 to 45 seconds, average 10.7 seconds; oil of thyme, 5 to 50 seconds, average 10.2 seconds; oil of wintergreen, 5 to 60 seconds, average 18 seconds. Two failed to respond to this odor. Leaves and stems of pennyroyal, 5 to 60 seconds, average 29.2 seconds. Seven failed to respond to this odor. Leaves and stems of spearmint, 5 to 60 seconds, average 24.7 seconds. Four failed to respond to this odor. Parts of decayed beetles, 5 to 30 seconds, average 13.4 seconds. The general average reaction time of the

25 beetles tested to all six odors is 17.7 seconds. These mutilated insects lived from 2 to 21 days with 9 days as an average. All the time they were confined in the small box, they drank, ate, and buried in the earth normally, but many times one was seen biting the soft dorsal portion of the abdomen of another. With the elytra and wings removed, the abdomens were unprotected and many of them shrank considerably in size before the beetles died. Some of these beetles were certainly killed on account of the dorsal sides of their abdomens being bitten, because nearly every one found dead had been entirely eaten except the chitinous parts. In the other two boxes as already mentioned, only occasionally was a dead beetle found that had been eaten.

(*c*) *Effects with Elytra and Wings Pulled Off and Pores on Legs Covered with Vaseline.*

The elytra and wings of 18 *Harpalus pennsylvanica* were pulled off at their articulations. Four days later the trochanters, femurs and proximal ends of the tibiæ of these mutilated beetles were covered with a vaseline-beeswax mixture, consisting of three fourths yellow commercial vaseline and one fourth beeswax. An hour after the legs had been vaselined, the beetles were placed singly into the experimental cases and were tested with the six odors as usual. Most of them were comparatively quiet, but a few were extremely restless. Their responses to odors were not pronounced and were slow, otherwise they were similar to those of unmutilated beetles.

Their reaction times are as follows: Oil of peppermint, 3 to 60 seconds, average 19.5 seconds. Three failed to respond to this odor. Oil of thyme, 3 to 60 seconds, average 12.5 seconds. Two failed to respond to this odor. Oil of wintergreen, 3 to 60 seconds, average 18.7 seconds. Four failed to respond to this odor. Leaves and stems of pennyroyal, 5 to 60 seconds, average 38.6 seconds. Nine failed to respond to this odor. Leaves and stems of spearmint, 3 to 60 seconds, average 32.9 seconds. Seven failed to respond to this odor. Parts of decayed beetles, 4 to 60 seconds, average 22.1 seconds. Two failed to respond to this odor. The general average reaction time of the 18 beetles tested to all six odors is 24.1 seconds. Confined in a box similar

to the other three already mentioned, these mutilated beetles drank, ate and buried in the earth normally, but they were less active than unmutilated ones. It was common to see them biting the dorsal sides of the abdomens. Before they died several of their abdomens had shrunk considerably in size. When found dead several of them had been entirely eaten except the chitinous parts. Counting from the time the elytra and wings were pulled off, they lived from 5 to 21 days with 10 days as an average.

THE OLFACTORY SENSE OF *Harpalus caliginosus*.

Eight ground beetles (*Harpalus caliginosus*) were caught under flat stones. They were tested with the odors from only the three essential oils. In behavior, they were comparatively quiet. When tested, many of them moved away quickly; a few vibrated the antennæ, and a few moved their legs.

Their reaction times are as follows: Oil of peppermint, 2 to 10 seconds, average 4.4 seconds; oil of thyme, 2 to 8 seconds, average 4.1 seconds; oil of wintergreen, 2 to 8 seconds, average 4.1 seconds. The general average reaction time to all three odors is 4.2 seconds. The antennæ of these beetles were pulled off and the insects were then kept in a small box containing earth in the table drawer.

(*a*) *Effects with Antennæ Pulled Off.*

Eight days after the antennæ of the eight preceding *Harpalus caliginosus* had been pulled off, the remaining six live ones were again tested with the same odors in the usual way. Their responses were similar to those given before they were mutilated, but were not so pronounced. When tested with the oil of thyme, one beetle rubbed a hind leg on an elytron for a half minute.

Their reaction times are as follows: Oil of peppermint, 3 to 25 seconds, average 12.5 seconds; oil of thyme, 4 to 60 seconds, average 14.3 seconds. One failed to respond to this odor. Oil of wintergreen, 10 to 35 seconds, average 22.5 seconds. The general average reaction time to all three odors is 16.4 seconds. These mutilated beetles were quite inactive and sometimes scarcely moved when touched with a pencil. They did not eat as greedily as before being mutilated. They lived from 2 to 65 days with 18 days as an average.

COCCINELLIDÆ.

The Olfactory Sense of *Epilachna borealis*.

Many lady beetles (*Epilachna borealis*) were caught on pumpkin vines in the corn field. When brought to the laboratory, they were put into a large glass jar near a window. The jar was 11 inches tall and 9 inches in diameter. It was covered with cheese-cloth. Since this lady beetle feeds upon the leaves of pumpkin and of allied plants, several pumpkin leaves were put into a wide-mouthed bottle containing water. The bottle with contents was then put into the jar. The beetles soon found the leaves and from that time on, they appeared "at home" as much as they do in corn fields on pumpkin leaves. They were regularly provided with a fresh supply of food. Occasionally they were seen copulating.

On the following day after being caught, 18 of them were removed from the jar and were put singly into the experimental cases. When mechanically irritated they draw in the antennæ and legs, usually eject a small drop of yellowish liquid from each femoro-tibial articulation, and feign death. They may lie apparently lifeless for several moments and when tested with odors they may or may not respond. Owing to this peculiar behavior, they were unsatisfactory to experiment with and their average reaction times are slower than might be expected. They were extremely quiet and when tested they generally moved away slowly. They often vibrated the antennæ and mouth parts, and sometimes the legs.

Their reaction times to the odors from the three essential oils are as follows: Oil of peppermint, 2 to 55 seconds, average 12.4 seconds; oil of thyme, 2 to 20 seconds, average 6.8 seconds; oil of wintergreen, 3 to 60 seconds, average 22.2 seconds. Three failed to respond to this odor. The general average reaction time to all three odors is 13.8 seconds. Sixteen of these insects were mutilated for other experiments. The seventeenth lived only 3 days and the eighteenth is still living at this writing (Jan. 15).

(*a*) *Effects with Antennæ Pulled Off.*

The antennæ of 25 *Epilachna borealis*, just caught, were pulled off at their bases. A small drop of yellowish blood exuded from

each wound. On the following day the beetles were tested with odors. As a rule they were so inactive that they appeared lifeless. If touched while moving they feigned death and remained inactive for several moments. When tested with odors most of them worked the mouth parts; some moved away slowly; a few vibrated one or more legs, and some failed to respond.

Their reaction times to the odors from the three essential oils are as follows: Oil of peppermint, 2 to 60 seconds, average 18.6 seconds. Three failed to respond to this odor. Oil of thyme, 2 to 60 seconds, average 38.7 seconds. Fourteen failed to respond to this odor. Oil of wintergreen, 3 to 60 seconds, average 35.1 seconds. The general average reaction time to all three odors is 30.8 seconds. Up to the time of this writing (Jan. 15), 15 of these mutilated beetles have died. They lived from 1 to 96 days with 22 days as an average.

(*b*) *Effects with Elytra and Wings Pulled Off.*

The elytra and wings of 10 *Epilachna borealis* were pulled off at their articulations. A small drop of yellowish blood exuded from each wound. A liquid of the same color is also present throughout the elytra and in the veins of the wings. On the second day after being mutilated, the four remaining live beetles were tested as usual. They were very quiet, but appeared normal in all respects except they responded to odors more slowly than unmutilated ones.

Their reaction times to the odors from the three essential oils are as follows: Oil of peppermint, 10 to 60 seconds, average 25 seconds. One failed to respond to this odor. Oil of thyme, 5 to 60 seconds, average 33.5 seconds. Two failed to respond to this odor. Oil of wintergreen, 7 to 60 seconds, average 35.5 seconds. Two failed to respond to this odor. The general average reaction time to all three odors is 31.3 seconds. Up to the time of this writing (Jan. 15), 1 of these beetles has died. Counting the 7 mutilated beetles that died, they lived from 2 to 3 days with 2 days as an average.

TELEPHORIDÆ.

THE OLFACTORY SENSE OF *Chaulcognathus pennsylvanica*.

Many fireflies (*Chaulcognathus pennsylvanica*) were caught on goldenrod (*Solidago*). They were put into a cage 20 inches long, 16 inches tall and 12 inches wide. The sides and top of the cage were cheesecloth while the ends and bottom were wood. The cage was kept in the light near a window and a fresh supply of goldenrod was constantly kept in the cage. On the goldenrod in the cage, these insects appeared quite "at home." Twenty-five of them were tested with the odors from the three essential oils. When tested most of them moved away quickly; a few vibrated antennæ; a few vibrated legs, and a few arose slowly. They were extremely restless at all times. In the cage they copulated as freely as they do out-of-doors.

Their reaction times are as follows: Oil of peppermint, 2 to 12 seconds, average 2.6 seconds; oil of thyme, 2 to 10 seconds, average 3 seconds; oil of wintergreen, 2 to 10 seconds, average 3 seconds. The general average reaction time to all three odors is 2.8 seconds. They lived from 3 to 7 days with 3.2 days as an average.

(*a*) *Effects with Antennæ Pulled Off.*

The antennæ of 27 *Chaulcognathus pennsylvanica* were pulled off at their bases. A day later only three were alive. When tested these three responded as promptly as unmutilated ones. The general average reaction time to the odors from the three essential oils is 2.8 seconds. Counting all 27 beetles, they lived from 1 to 5 days with 1.3 days as an average.

LUCANIDÆ.

THE OLFACTORY SENSE OF *Passalus cornutus*.

Four stag beetles (*Passalus cornutus*) were removed from rotten stumps. While being tested with odors they were comparatively quiet and responded promptly. Their most common response was to draw in the antennæ and to move away slowly. The general average reaction time to all six odors is 3.2 seconds. The antennæ were pulled off at their bases. A small drop of

blood exuded from each wound. The beetles were kept in a small box filled with moist rotten wood.

(*a*) *Effects with Antennæ Pulled Off.*

Two days after pulling off the antennæ, the four preceding mutilated beetles were again tested with the same odors. They were more quiet than before being mutilated. Their responses were just as prompt but were less pronounced than before they were mutilated. Their most common response was to work the mouth parts and to move away slowly. The general average reaction time to all six odors is 3.3 seconds. They lived from 4 to 20 days with 12.5 days as an average.

SCARABÆIDÆ.

THE OLFACTORY SENSE OF *Cotinis nitida.*

One lamellicorn beetle (*Cotinis nitida*) was tested with the six odors. The most common response was to stretch out its head, and to move its antennæ and front legs. Once it drew in the antennæ and moved the front legs. The average reaction time is 8 seconds. The antennæ were pulled off at their bases. A small drop of blood exuded from each wound.

(*a*) *Effects with Antennæ Pulled Off.*

A day after pulling off the antennæ, the preceding *Cotinis nitida* was again tested with the same odors. It responded as promptly as before being mutilated. The most common response was to work the mouth parts and to move away slowly. The average reaction time is 8.3 seconds. It lived 12 days after being mutilated.

THE OLFACTORY SENSE OF *Euphoria sepulchralis.*

Five lamellicorn beetles (*Euphoria sepulchralis*) were caught on goldenrod (*Solidago*). While being tested with the odors from the three essential oils, they were extremely restless. They generally moved away slowly and drew in the antennæ when tested with an odor. The general average reaction time is 3.6 seconds. After the antennæ had been pulled off at their bases, the beetles were put into the cage described on page 436.

(*a*) *Effects with Antennæ Pulled Off.*

A day later the five preceding mutilated insects were again tested with the same odors. They were quiet and their responses were similar to those before being mutilated, except, of course, there were no antennal movements. The general average reaction time is 4.3 seconds. These beetles lived from 9 to 42 days with 20 days as an average after being mutilated.

CERAMBYCIDÆ.

THE OLFACTORY SENSE OF *Cyllene robiniæ.*

Eighteen wood-boring beetles (*Cyllene robiniæ*) were caught on goldenrod (*Solidago*). While being tested with the odors from the three essential oils, they were extremely restless. When tested, most of them moved away quickly; a few arose quickly, and a few vibrated the antennæ. The general average reaction time is 5.4 seconds. These beetles were confined in the cage described on page 436. They were regularly given a fresh supply of goldenrod. They seemed "at home" and copulated as freely in the cage as they do out-of-doors. They lived from 1 to 17 days with 10.4 days as an average.

(*a*) *Effects with Antennæ Pulled Off.*

Eighteen more *Cyllene robiniæ* were collected from goldenrod. Their antennæ were pulled off at the bases. A small drop of blood exuded from each wound. These beetles were placed into the cage with the unmutilated ones. Two days later the 15 remaining live ones were tested with the odors from the essential oils. They were very quiet and their responses were similar to those of unmutilated individuals, except as a rule they were more prompt. The general average reaction time is 3 seconds. In the cage it was common to see the unmutilated and antennæless cerambycids copulating. The former were very active and flew out of the cage whenever the door was opened, but the latter seldom flew and they were not so active. The mutilated ones lived from 1 to 11 days with 5 days as an average.

(*b*) *Effects with Elytra and Wings Pulled Off.*

Eighteen more *Cyllene robiniæ* were collected. Their elytra and wings were pulled off at the articulations. A small drop

of blood always exuded from each wound caused by the elytron being pulled off, but only occasionally was blood seen where a wing had been pulled off. A day later when tested with the odors from the three essential oils, these beetles were comparatively quiet and they appeared normal in all respects except in their slowness in responding to odors.

Their reaction times are as follows: Oil of peppermint, 2 to 30 seconds, average 7.1 seconds; oil of thyme, 3 to 20 seconds, average 8.9 seconds; oil of wintergreen 3 to 55 seconds, average 13.4 seconds. The general average reaction time to all three odors is 9.8 seconds. In the cage with the other beetles, these mutilated ones were as active as the unmutilated cerambycids and they were often seen copulating with each other, and with the unmutilated and antennæless ones. They lived from 1 to 11 days with 4.2 days as an average.

CHRYSOMELIDÆ.

THE OLFACTORY SENSE OF *Leptinotarsa 10-lineata*.

Forty-five Colorado potato beetles (*Leptinotarsa 10-lineata*) were collected in a potato patch near the laboratory. While 25 of them were being tested with the six odors, they were comparatively quiet as a rule, but five were so restless that they were discarded and others were used. Their responses were similar to those of *Harpalus pennsylvanica*, described on page 429.

Their reaction times are as follows: Oil of peppermint, 2 to 7 seconds, average 3.3 seconds; oil of thyme, 2 to 5 seconds, average 3.1 seconds; oil of wintergreen, 2 to 12 seconds, average 5 seconds; leaves and stems of pennyroyal, 4 to 60 seconds, average 26.7 seconds. Six failed to respond to this odor. Leaves and stems of spearmint, 2 to 60 seconds, average 25.6 seconds. Seven failed to respond to this odor. Parts of decayed beetles, 5 to 60 seconds, average 27.9 seconds. Seven failed to respond to this odor. The general average reaction time of the 25 beetles tested to all six odors is 15.4 seconds. These insects were confined in a cage in the light near a window. This cage is 30 inches long, 30 inches high and 4½ inches wide. All six sides are wire-screen. A fresh supply of potato plant leaves was constantly kept in the cage. The beetles confined in this cage on the potato plant

leaves appeared "at home" just as much as they do in potato patches. They ate the leaves, copulated and laid eggs as usual. Up to the time of this writing (Jan. 15), 28 of the 45 beetles have died. These lived from 14 to 151 days with 69 days as an average.

(*a*) *Effects with Antennæ Pulled Off.*

Twenty-nine more potato beetles were collected from the potato patch. Their antennæ were pulled off at the bases. A small drop of blood exuded from each wound. These insects were put into the wire-screen cage with the unmutilated ones. Two days later the 23 remaining live ones were tested with only the odors from the three essential oils. All of these beetles were quite inactive and three failed to respond when tested. These three also failed to respond when touched with a pencil. For this reason they were discarded. The general average reaction time of the 20 beetles tested is 3.5 seconds. As a rule these mutilated insects appeared normal in all respects several days after having the antennæ pulled off, because they ate, copulated and were as active as ever. They lived from 2 to 140 days with 38 days as an average.

(*b*) *Effects with Elytra Pulled Off and Wings Cut Off.*

Thirty-one more potato beetles were collected. Their elytra were pulled off at the articulations and the wings were cut off as closely as possible to the articulations. A small drop of reddish or yellowish blood exuded from each wound. The heavy veins, extending from the base of the wing to where the wing folds, contain most of the blood found in these wings. The elytra are also filled with blood. The amount of blood in them gradually diminishes from the base to the distal end. A day after being mutilated 25 of these insects were tested with the six odors. They were apparently normal in all respects except in their slowness in responding to odors. They were as active as unmutilated ones and eight were extremely restless. Their responses were similar to those of unmutilated beetles, except they were not pronounced.

Their reaction times are as follows: Oil of peppermint, 2 to 40

seconds, average 7.8 seconds; oil of thyme, 2 to 15 seconds, average 4.8 seconds; oil of wintergreen, 3 to 60 seconds, average 21.1 seconds. Five failed to respond to this odor. Leaves and stems of pennyroyal, 5 to 60 seconds, average 32.2 seconds. Ten failed to respond to this odor. Leaves and stems of spearmint, 3 to 60 seconds, average 29.8 seconds. Eight failed to respond to this odor. Parts of decayed beetles, 3 to 60 seconds, average 30.4 seconds. Eight failed to respond to this odor. The general average reaction time of the 25 beetles tested to the six odors is 22.7 seconds. In the wire-screen cage with the other potato beetles already tested, these mutilated ones appeared normal, because they ate normally and copulated as much as usual. Since the soft dorsal sides of their abdomens were unprotected, many of them soon began to sink, so that by the time a beetle died, the abdomen had shrunk to about one-fourth its original size. Up to the time of this writing (Jan. 15), 29 of these 31 mutilated insects have died. They lived from 3 to 140 days with 52 days as an average.

(c) *Effects with Elytra Pulled Off, Bases of Wings Glued and Pores on Legs Covered with Vaseline.*

Twenty-nine more potato beetles were collected. Their elytra were pulled off at the articulations. Two days later the upper surfaces of the bases of the wings of the 26 remaining live ones were covered with liquid glue. Since the olfactory pores extend a considerable distance from the base of the wing along the radial vein, the glue applied probably did not cover more than 90 per cent. of the pores on each wing. Three hours after applying the glue, the trochanters, femurs and proximal ends of the tibiæ of these beetles were covered with the vaseline-beeswax mixture. An hour still later the insects were tested with the six odors. They were as active as unmutilated ones and appeared normal in all respects except in their responses to odors. Their responses were never pronounced and seldom prompt.

Their reaction times are as follows: Oil of peppermint, 3 to 60 seconds, average 10.7 seconds. One failed to respond to this odor. Oil of thyme, 3 to 60 seconds, average 9 seconds. One failed to respond to this odor. Oil of wintergreen, 5 to 60 seconds,

average 35.9 seconds. Eleven did not respond to this odor. Leaves and stems of pennyroyal, 3 to 60 seconds, average 35.2 seconds. Twelve did not respond to this odor. Leaves and stems of spearmint, 5 to 60 seconds, average 42.6 seconds. Fourteen failed to respond to this odor. Parts of decayed beetles, 5 to 60 seconds, average 40.3 seconds. Fourteen failed to respond to this odor. The general average reaction time of the 26 beetles tested to the six odors is 29 seconds which is twice the reaction time of unmutilated potato beetles to the same odors. When the reaction times to the odors from only the three essential oils are considered, these mutilated insects responded only one fifth as rapidly as did the unmutilated ones. In the wire-screen cage with the other potato beetles already tested, they were apparently normal as long as they lived, because they ate and copulated as usual and were always as active as the unmutilated ones. Before they died their abdomens shrunk considerably in size. Up to the time of this writing (Jan. 15), 28 of the 29 have died. These lived from 2 to 151 days with 61 days as an average.

MELOIDÆ.

The Olfactory Sense of *Epicauta marginata*.

Twenty blister beetles (*Epicauta marginata*), commonly known as the "old-fashioned potato bugs," were caught on clematis. When mechanically irritated, they fold the antennæ and legs against the body, usually eject a small drop of amber-colored liquid from each femoro-tibial articulation, and feign death. On account of this behavior, they were unsatisfactory to experiment with. When put into the experimental cases, some of them lay apparently lifeless for almost a half day. In this state they never respond to any odor, and after becoming as active as usual, they may or may not respond to odors.

When tested with the odors from only the three essential oils, a general average reaction time of 13.9 seconds was obtained. Two of them failed to respond to each of the oils of peppermint and wintergreen. These insects were confined in the cage described on page 436. They were regularly provided with a fresh supply of clematis. In this cage on the clematis they seemed "at home," but they flew out at every opportunity.

They copulated as usual. They lived from 11 to 40 days with 27.6 days as an average.

(*a*) *Effects with Antennæ Cut Off.*

Eight more *Epicauta marginata* were collected. Their antennæ were cut off at the bases. A small drop of amber-colored blood exuded from each wound. Seven days later the two remaining live ones were tested with the odors from the three essential oils. The general average reaction time is 5 seconds. All these beetles were abnormal in behavior. They lived from 1 to 8 days with 3.4 days as an average.

(*b*) *Effects with Antennæ Pulled Off.*

The antennæ of 12 more *Epicauta marginata* were pulled off at their bases. A small drop of blood exuded from each wound. When tested with the odors from the essential oils three days later, the eight remaining live beetles gave a general reaction time of 5.9 seconds. They were less abnormal in behavior than those with the antennæ cut off. They lived from 2 to 13 days with 5.5 days as an average.

(*c*) *Effects with Elytra and Wings Pulled Off.*

The elytra and wings of nine *Epicauta marginata* were pulled off at their articulations. A small drop of blood exuded from each wound. When tested with the odors from the essential oils two days later, the seven remaining live beetles gave a general reaction time of 25.7 seconds. Two of them failed to respond to each of the oils of peppermint and wintergreen. These mutilated insects appeared normal in behavior and in confinement they copulated as usual. They lived from 2 to 14 days with 8 days as an average.

THE OLFACTORY SENSE OF *Epicauta pennsylvanica.*

Twenty-five blister beetles (*Epicauta pennsylvanica*) were caught on golden rod (*Solidago*). This species has the same habit of feigning death when mechanically irritated as has *Epicauta marginata*. When tested with the odors from the essential oils, they gave a general average reaction time of 11.5 seconds

which is only one-half as rapid as the reaction time of the same species devoid of antennæ. Three failed to respond to the oil of peppermint, one to the oil of thyme and two to the oil of wintergreen. A common response was to vibrate the legs. They were placed into the cage with the other species of blister beetles. They were regularly provided with a fresh supply of goldenrod. In the cage they appeared normal, and they copulated as much as usual. They lived from 2 to 25 days with 11.2 days as an average.

(*a*) *Effects with Antennæ Pulled Off.*

The antennæ of 30 *Epicauta pennsylvanica* were pulled off at their bases. When tested with the odors from the essential oils three days later, the 22 remaining live beetles gave a general reaction time of 5.3 seconds. They were only slightly abnormal in behavior. They lived from 2 to 25 days with 8.7 days as an average.

(*b*) *Effects with Elytra and Wings Pulled Off.*

The elytra and wings of 21 *Epicauta pennsylvanica* were pulled off at their articulations. A small drop of blood exuded from each wound. Blood was also seen in the distal ends of the elytra. When tested with the odors from the essential oils two days later, the 17 remaining live beetles gave a general reaction time of nine seconds. One of them failed to respond to the oils of thyme and wintergreen. These insects appeared normal in confinement with the other blister beetles. They copulated as usual. They lived from 1 to 33 days with 10.7 days as an average.

A summary of all the preceding experiments to determine the location of the olfactory organs in beetles is best presented in a tabulated form. The following table is such a summary. Since a comparison of the behavior of unmutilated and mutilated insects alone is not always a safe criterion for judging the general behavior of mutilated beetles, the behavoir of the mutilated beetles recorded in this table is based mostly upon a comparison of the longevities of unmutilated and mutilated individuals of the same species. A "+" after a figure in the last column means that all the insects used in the experiment have not yet died. The longevity is based only on those that have died up to the time of this writing (Jan. 15).

TABLE II.

SUMMARY OF EXPERIMENTS TO DETERMINE THE LOCATION OF THE OLFACTORY ORGANS IN COLEOPTERA.

Species.	Experiment and Behavior of Insects Tested.	Average Reaction Time. For Three Odors.	Average Reaction Time. For Six Odors.	No. of Individuals Tested.	Average Length of Life in Captivity.
		Sec.	Sec.		Days.
Harpalus pennsylvanica.	Unmutilated. Normal in behavior	9.5	16.5	25	61.0+
	Antennæ pulled off. Normal in behavior	5.1	16.1	25	58.0+
	Elytra and wings pulled off. Slightly abnormal in behavior.	13.0	17.7	25	9.0
	Elytra and wings pulled off and pores on legs covered with vaseline. Slightly abnormal in behavior	16.9	24.1	18	10.0
Harpalus caliginosus.	Unmutilated. Normal in behavior	4.2		8	Used below
	Antennæ pulled off. Slightly abnormal in behavior	16.4		6	18.0
Epilachna borealis.	Unmutilated. Normal in behavior	13.8		18	Used below
	Antennæ pulled off. Slightly abnormal in behavior	30.8		25	22.0+
	Elytra and wings pulled off. Slightly abnormal in behavior	31.3		4	2.0+
Chaulcognathus pennsylvanica.	Unmutilated. Normal in behavior	2.8		25	3.2
	Antennæ pulled off. Slightly abnormal in behavior	2.8		3	1.3
Passalus cornutus.	Unmutilated. Normal in behavior	3.0	3.2	4	Used below
	Antennæ pulled off. Slightly abnormal in behavior	3.0	3.3	4	12.5
Cotinis nitida	Unmutilated. Normal in behavior	5.0	8.0	1	Used below
	Antennæ pulled off. Normal in behavior	5.6	8.3	1	12.0
Euphoria sepulchralis.	Unmutilated. Normal in behavior	3.6		5	Used below
	Antennæ pulled off. Normal in behavior	4.3		5	20.0
Cyllene robiniæ	Unmutilated. Normal in behavior	5.4		18	10.4
	Antennæ pulled off. Slightly abnormal in behavior	3.0		15	5.0
	Elytra and wings pulled off. Slightly abnormal in behavior	9.8		18	4.2

Species.	Experiment and Behavior of Insects Tested.	Average Reaction Time. For Three Odors.	Average Reaction Time. For Six Odors.	No. of Individuals Tested.	Average Length of Life in Captivity.
		Sec.	Sec.		Days.
Leptinotarsa 10-lineata.	Unmutilated. Normal in behavior	3.8	15.4	25	69.0+
	Antennæ pulled off. Normal in behavior	3.5		20	38.0
	Elytra pulled off and wings cut off. Normal in behavior	11.2	22.7	25	52.0+
	Elytra pulled off, bases of wings glued and pores on legs covered with vaseline. Normal in behavior	18.5	29.0	26	61.0+
Epicauta marginata.	Unmutilated. Normal in behavior	13.9		20	27.6
	Antennæ cut off. Considerably abnormal in behavior	5.0		2	3.4
	Antennæ pulled off. Slightly abnormal in behavior	5.9		8	5.5
	Elytra and wings pulled off. Slightly abnormal in behavior	25.7		7	8.0
Epicauta pennsylvanica.	Unmutilated. Normal in behavior	11.5		25	11.2
	Antennæ pulled off. Slightly abnormal in behavior	5.3		22	8.7
	Elytra and wings pulled off. Normal in behavior	9.0		17	10.7

A summary of the preceding table shows the following: After the antennæ were pulled off, four of the 11 species tested were normal and seven were slightly abnormal in behavior. After the elytra and wings were pulled off one species was normal while four were slightly abnormal in behavior. After the elytra were pulled off and the wings were cut off, the one species tested was normal in behavior. After the elytra and wings were pulled off and the pores on the legs were covered with vaseline, the one species tested was slightly abnormal in behavior. After the elytra were pulled off, the bases of the wings glued and the pores on the legs covered with vaseline, the one species tested was normal in behavior.

Four unmutilated species responded to odors more slowly than did the same species after the antennæ had been pulled off. This is explained by the fact that most beetles are more or less "timid"

for some time after being caught, and some feign death. As a rule the longer they are confined and the more they are handled, the more satisfactory they are to experiment with. Five species without antennæ responded to odors as promptly as did the same species unmutilated. Two species without antennæ responded to odors more slowly than did the same species unmutilated. Since these were abnormal in behavior and judging from the reaction times of the other nine species with antennæ pulled off, it is only reasonably to attribute the slow reaction times of these two species to their abnormal condition caused by the antennæ being pulled off. The six species so mutilated that most of their olfactory pores on the elytra and wings were prevented from functioning responded from two to five times more slowly than did the same species unmutilated or with the antennæ pulled off. The two species so mutilated that most of their olfactory pores on the elytra, wings and legs were prevented from functioning responded from two to six times more slowly than did the same species unmutilated or with the antennæ pulled off.

From all the preceding results, it seems that the antennæ do not carry any of the olfactory organs, while the olfactory pores found on the peduncles of the elytra, on the dorsal surfaces of the wings, on the trochanters, tibiæ, sometimes on the femurs and tarsi, and perhaps on the mouth appendages, are the true olfactory organs in beetles.

SUMMARY.

In making a comparative study of the olfactory pores in beetles, 50 species belonging to 47 genera and representing 34 families were used. A group of pores is always present on the peduncle of each elytron. It lies on the dorsal side of the well-exposed radial plate. The number of pores on a pair of elytra varies from 12 to 310. As a rule, the more pores in the group the smaller they are and the closer they are together.

Of the 47 winged species examined, 11 have only one group of pores on each wing, 21 have two groups on each wing, 12 have three groups on each wing, and 3 have four groups on each wing. These groups are always located on the dorsal surface. Only occasionally are a few scattered pores found on the ventral side

of a wing. When one or two groups are present, they lie on the radius. When three groups are present, all three may lie on the radius, or two may lie on the radius and the third on the media. When four groups are present, one lies on the subcosta, two on the radius and one on the media. The largest group on the radius usually extends nearly all the way to the fold of the wing and sometimes all the distance to the fold. The number of pores on a pair of wings varies from 130 to 982.

There are usually two groups of pores at the proximal end of each trochanter. Sometimes a pore is found at the proximal end of the femur. It is common to find a few pores at the proximal end of each tibia; and sometimes pores are found in the tibial spines and on the tarsi. The number of pores on all six legs varies from 49 to 341.

In regard to water beetles, the better the legs are adapted for locomotion in water, the fewer pores they have. The smallest winged species (*Coxelus*) examined has 273 pores, which is the smallest number of all the species, and the largest species (*Orthosoma*) has 1,268 pores which is the largest number of all the species examined. The apterous species have more pores on the legs than usual. As a rule, the smaller the species, the fewer its pores and the larger they are, comparatively speaking. As a rule, there are no generic and specific differences, except variations in number of pores, the amount of variation depending on the sizes of the individuals compared. There are no individual and sexual differences other than slight variations in number of pores.

The pore apertures or pits are round, oblong, slitlike or club-shaped. On the elytra and wings they are always round or oblong. On the legs they have all four of the enumerated shapes.

The spindle-shaped sense cells of most beetles lie in the lumens of the appendages outside the pore cavities, but in the legs of *Orthosoma* the sense cells lie inside the pore cavities. A small chitinous cone is always present. It is formed by the hypodermal cell at the mouth of the pore after the insect has emerged from the last pupal stage, and at the same time when the chitinous integument is being considerably thickened. The sense cells are fully developed when the insect emerges into the imago stage. The sense fiber pierces the cone and the layer of chitin between

the pore aperture and cone, and it enters the bottom of the pore aperture or pit where its peripheral end comes into direct contact with the outside air. In Hymenoptera the sense fibers enter the pore apertures which are almost on a level with the external surface of the chitin. In Coleoptera, with a few exceptions, the sense fibers enter the bottoms of pits which lie in the chitin one third (at time of emerging into imago stage) the distance from the external surface. In the legs of the lady beetle, *Epilachna borealis*, instead of the chitin which surrounds the pore apertures being depressed, it is elevated so that the pore apertures lie in the center of domes above the general surface of the legs. In the legs of the blister beetles, *Epicauta marginata* and *E. pennsylvanica*, the pore apertures lie on a level with the surface of the legs. In the legs of the potato beetle, the pore apertures lie at the bottoms of shallow pits. All four preceding species have hypodermal gland pores over the entire body, except the wings. These pores in the lady beetle are perhaps the most highly developed. They lie on all sides and even among the olfactory pores on the legs. In the other three species they are less highly developed on the legs near the olfactory pores and none is found very close to an olfactory pore. This correlation between the hypodermal gland pores and the olfactory pores is certainly a means of preventing the secretion from the gland cells from running into the pore apertures.

A large nerve and a large trachea run into each elytron and wing. In the peduncle of the elytron they run through the radial plate just beneath the group of olfactory pores. Branches from the nerve are given off which connect with the sense cells. The large nerve and trachea passing into the wing soon divide so that a smaller nerve and a smaller trachea run through each main nerve. The largest trachea passes through the subcosta, and the largest nerves pass through the veins carrying the olfactory pores. These nerves give off branches which connect with the sense cells. The sense cells wherever found are always surrounded by blood.

In the experiments to determine the location of the olfactory organs, 434 individuals were tested. These belonged to 11 species representing 8 families. After the antennæ were pulled

off, 4 of the 11 species tested were normal and 7 were slightly abnormal in behavior. After the elytra and wings were pulled off 1 species was normal while 4 were slightly abnormal in behavior. After the elytra were pulled off and the wings were cut off, the 1 species tested was normal in behavior. After the elytra and wings were pulled off and the pores on the legs were covered with vaseline, the 1 species tested was slightly abnormal in behavior. After the elytra were pulled off, the bases of the wings glued and the pores on the legs covered with vaseline, the 1 species tested was normal in behavior.

Four unmutilated species responded to odors more slowly than did the same species after the antennæ had been pulled off. This is explained by the fact that most beetles are more or less "timid" for some time after being caught, and some feign death. As a rule, the longer they are confined and the more they are handled, the more satisfactory they are to experiment with. Five species without antennæ responded to odors as promptly as did the same species unmutilated. Two species without antennæ responded to odors more slowly than did the same species unmutilated. Since these were abnormal in behavior and judging from the reaction times of the other 9 species with antennæ pulled off, it is only reasonable to attribute the slow reaction times of these two species to their abnormal condition caused by the antennæ being pulled off. The 6 species so mutilated that most of their olfactory pores on the elytra and wings were prevented from functioning responded from 2 to 5 times more slowly than did the same species unmutilated or with the antennæ pulled off. The two species so mutilated that most of their olfactory pores on the elytra, wings and legs were prevented from functioning responded from 2 to 6 times more slowly than did the same species unmutilated or with the antennæ pulled off.

From all the preceding results, it seems that the antennæ do not carry any of the olfactory organs, while the olfactory pores found on the peduncles of the elytra, on the dorsal surfaces of the wings, on the trochanters, tibiæ, sometimes on the femurs and tarsi, and perhaps on the mouth appendages, are the true olfactory organs in beetles.

DISCUSSION.

Since the writer ('14*c*) has already written a complete review of all the literature available concerning the sense of smell in insects, only a brief discussion is necessary in this paper.

Hicks ('57) says that the olfactory pores in Coleoptera are arranged in long rows along the subcostal nerves. The same author ('59) states that in Coleoptera these organs are highly developed and occur in numerous groups on the subcostal vein, mostly at the widest part, but are also scattered along it to the fold of the wing. In *Carabus* they are found on veins other than the subcostal. In many beetles the pore is overarched by a hair, which probably protects the organ. He could distinguish no sexual differences in these organs, except the pores are slightly larger in the females, due to their greater size. Hicks ('60) first found the olfactory pores on the legs of beetles. The present writer has never seen a hair overarching an olfactory pore.

Hochreuther ('12) seems to be the first to study the internal anatomy of the olfactory pores in beetles. Since he used only *Dytiscus marginalis* and perhaps because he did not have enough sections through these organs, he failed to understand their anatomy. He states that each dome-shaped organ is located at the bottom of a chitinous flask, the mouth of which communicates with the exterior. Instead of the peripheral end of the sense fiber coming into direct contact with the air in the flask, it apparently stops just beneath the chitinous dome at the top of the organ. His terminal strand (Terminalstrand) may be the same as the hypodermal secretion forming the cone described by the writer. Hochreuther found a few of these dome-shaped organs on the epicranium near the margin of the eyes, nine on the proximal end of the first antennal segment, two on the distal end of the second antennal segment, a few on the dorsal side of the labrum, a very few on the dorsal side of the mandible, several on each maxilla, about 18 on the first four segments of the front legs, about 10 on the first three segments of the middle legs, and a few on the trochanters of the hind legs. He evidently did not examine the wings. Thus according to Hochreuther these organs are rather widely distributed. Since the peripheral ends of the sense fibers do not come into contact with the outside air,

but connect with the tops of the domes, he suggests that they receive some kind of mechanical stimuli, although he performed no experiments to determine their function.

Lehr ('14), resuming the search for sense organs in *Dytiscus marginalis* where left off by Hochreuther, found dome-shaped organs on the elytra and wings. He found three main groups in identically the same places as described by the present writer. The number of pores in the group on the elytron varies from 130 to 150. The two main groups on the radius (his subcosta) of the wing are large, but he did not count the pores in them. He found a fourth group, consisting of about 30 pores, on the ventral side of the costa near the base of the wing. He also found a few scattered pores on the dorsal side of the costa just distal to the fold of the wing, a few on the second cubitus, and a few irregularly scattered along the full length of the media. Lehr has described the anatomy of these organs almost identically as seen by the present writer, but it seems that he has not correctly interpreted some of the structures. He seems to think that each sense cell is surrounded by another cell, but the latter cell is perhaps nothing more than coagulated blood and the portion of it extending into the pore is certainly a hypodermal secretion forming the cone as described in the preceding pages. His neurilemma nuclei are perhaps hypodermal nuclei. He is able to trace the sense fiber through the cone, but he has not recognized the small opening through the dome. This is not surprising, because the pores in the wings as so small that the openings or pore apertures are never noticed unless first seen in the largest pores in the legs or mouth parts. In the thinnest sections, the chitin forming the dome is so thick as compared to the diameter of the pore aperture that the aperture appears only as a streak slightly lighter than the other chitin in the dome. Lehr has nothing to say about the physiology of these organs.

In experimenting with mutilated beetles, Hauser ('80) seems to be the only one who has taken their longevity into consideration. And even he has not kept an accurate record of their behavior and longevity. He claims to have studied the behavior of beetles before and after the removal of the antennæ. When the antennæ were removed he ascertained that many beetles

soon became sick and died, while others lived thereafter for many days. When tested with odors, most of the beetles without antennæ failed to respond, but Hauser states that *Carabus*, *Melolontha* and *Silpha* still responded to odors, although more slowly.

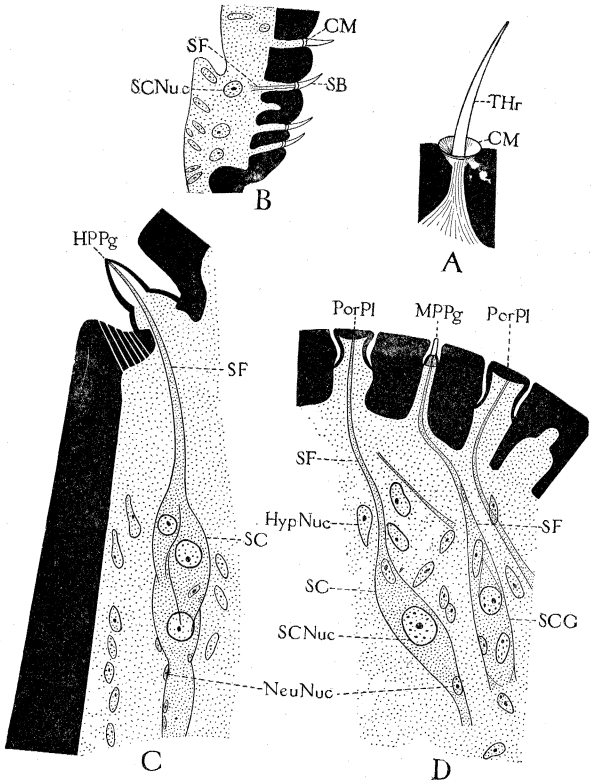

FIG. 3. Antennal organs of the water beetle, *Dytiscus marginalis*, copied from Hochreuther (1912). *A*, small tactile hair (Sinneshaar) from first segment of antenna, total preparation (Fig. 1 from Hochreuther), × 330; *B*, portion of Fig. 12 from Hochreuther, showing four small sense bristles (Sinnesborsten) from proximal end of second segment of antenna, × 265; *C*, longitudinal section (Fig. 48 from Hochreuther) through a hollow pit peg (hohlen Grubenkegel), × 470; *D*, longitudinal section through a small massive pit peg (massiven, grubenständigen Zapfen) and two pore plates (kelchförmige Organe), × 590. This drawing is a combination of Figs. 32 and 58 from Hochreuther. Only the pore plates (*PorPl*) are taken from Fig. 58. Hochreuther gives a drawing of only one perfect pore-plate organ, or cup-shaped organ, and it is from the maxillary palpus. *CM*, cup-shaped membrane; *HPPg*, hollow pit peg; *MPPg*, massive pit peg; *PorPl*, pore plate; *SB*, sense bristle; *THr*, tactile hair. See page 456 for other abbreviations.

For the purpose of judging whether the antennal organs are better adapted anatomically than the olfactory pores for receiving odor stimuli, the former organs (Text-fig. 3, p. 453), of *Dytiscus marginalis* have been copied from Hochreuther ('12). This work of Hochreuther is a comprehensive study of the morphology of all the chitinous sense organs of *Dytiscus*. Since it is perhaps the latest and certainly the best study on the antennal organs of beetles, these organs shall be briefly described.

Each of the 11 segments in the antenna of *Dytiscus* carries a number of sense organs. The farther from the base of the antenna the more numerous they are. The distal half of the antenna is covered abundantly with sense organs, while the proximal half is sparingly covered with them. The first and second segments are well provided with slender tactile hairs (Text-fig. 3*A*, *THr*) which have been called *Sensilla trichodea* by Schenk. These hairs are also found on all the other appendages and even on the head, thorax and abdomen. Two groups of sense bristles (Text-fig. 3*B*, *SB*), called *Sensilla chætica* by Schenk, lie at the proximal end of the second segment. These hairs are also common on most of the other appendages, on the head, thorax and abdomen. All segments, except the first one, are well provided with small massive pit pegs of the thick-walled type (Text-fig. 3*D*, *MPPg*). All segments, except the first one, are only sparingly provided with a second type of pit pegs. This one is the hollow or thin-walled type (Text-fig. 3*C*, *HPPg*). Only about six of these were found on each segment. Besides being found on the antennæ, both types of pit pegs are common on all the mouth parts, on the mesothorax, around the spiracles, on all the legs, and on the sexual apparatus. Pit pegs have been called *Sensilla coelloconica* by Schenk. All segments, except the first two, are abundantly supplied with the cup-shaped or pore-plate organs (Text-fig. 3*D*, *PorPl*). For both antennæ they are estimated between 4,500 and 5,000. These organs are also common on the palpus of the first maxilla. They were first studied by Nagel on the antennæ and maxillary palpi of Dytiscidæ. In the honey bee Schenk has called them *Sensilla placodea*. Of the five antennal organs of *Dytiscus*, only the hollow pit pegs are regarded by Hochreuther as probably ol-

factory in function. If they really act as olfactory organs, then the mouth parts, thorax, legs and sexual organs must aid in receiving odor stimuli. Hochreuther considers the antennæ more important as appendages for carrying organs for receiving mechanical stimuli rather than those receiving chemical stimuli.

According to various authors the antennal organs of different beetles vary only slightly. The antennal organs of *Dytiscus* are also similar to those of the honey bee. In both of these insects the tactile hairs are of the same type. The Forel flasks and pit pegs of the honey bee are two types of pit pegs which are perhaps rudimentary, because the tips of the hairs do not come to the exterior of the chitin. The massive pit pegs, hollow pit pegs, and the sense bristles of *Dytiscus* are certainly nothing more than three types of tactile hairs. The hollow pit pegs compare closely with the pegs of the honey bee, except the pegs have thinner chitin at the tips. This is probably on account of more acute sense of touch in the honey bee. The pore-plate organs of the honey bee and the cup-shaped organs of *Dytiscus* are also quite similar.

One or more of the antennal organs of every insect studied have been called olfactory organs, and it is possible that most of these organs may be found on other appendages, besides the antennæ, as already seen in *Dytiscus*.

In conclusion it seems beyond a doubt that none of the antennal organs of beetles shown in Text-fig. 3 serves as an olfactory organ, and that the olfactory pores are well adapted anatomically for receiving odor stimuli, because the peripheral ends of their sense fibers come into direct contact with the external air.

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EXPLANATION OF PLATES I. AND II.

All figures including Text-figs. 1 and 2 are from camera lucida drawings made at the base of the microscope. Figures 1 to 8 inclusive and 22 on the plates are enlarged 465 diameters. All the remaining figures on the plates, except the diagrams 28, 29 and 31, are enlarged 580 diameters.

ABBREVIATIONS.

BlSin.....blood sinus.
BM.....basal margin of elytron.
C.....costa.
Ch.....chitin.
Ch$_1$.....chitin formed before insect emerges into imago stage.
Ch$_2$.....chitin formed after insect emerges from last pupal stage.
ChM.....chitinous membrane of pore plate.
CM.....cup-shaped membrane of tactile hair on antenna.
Con.....chitinous cone.
ConT.....connective tissue.
ConTNuc....nucleus of connective tissue.
Cx.....coxa.
F.....femur.
Fl.....flange of olfactory pore.
Fo.....where wing folds.
GlC.....gland cell.
HPPg.....hollow pit peg on antenna.
Hr.....hair.
Hyp.....hypodermis.
Hyp$_1$.....membrane resembling hypodermis which divides the lumen of proximal end of the tibia of *Epilachna* into two chambers.
HypC.....hypodermal cell.
HypNuc.....hypodermal nucleus.
HypS.....hypodermal secretion.
M.....muscle.
MD.....muscle disk.
Me.....media.
Mo.....mouth of pore.

MPPg.....small massive pit peg on antenna.
N.....nerve.
NB.....nerve branch.
Neu.....neurilemma.
NeuNuc.....nucleus of neurilemma.
NeurNuc....neuroglia nucleus.
P.....pit of pore.
PorAp.....pore aperture.
PorGl.....pore of gland.
PorHr.....pore of hair.
PorPl.....pore plate on antenna.
Por.....pore of olfactory organ.
PorW.....pore wall.
PorWGl.....pore wall of gland.
PorWHr.....pore wall of hair.
R.....radius.
RP.....radial plate.
SB.....small sense bristle on antenna.
SC.....sense cell.
SC_1.....sense cell of tactile hair
SCG.....sense cell group.
SCNuc.....sense cell nucleus.
SCNucl.....sense cell nucleolus.
Sc.....subcosta.
ScH.....subcostal head.
SF.....sense fiber.
Tar.....tarsus.
Tb.....tibia.
TbSp.....tibial spine.
THr.....tactile hair.
Tr.....trachea.
TrNuc.....Nucleus of trachea.
Tro.....trochanter.
1 to 6.....groups Nos. 1 to 6 of the olfactory pores.
a.....location of scattered pores on ventral side of wing.
b and *c*.....location of scattered pores on tibia.

PLATE I.

FIG. 1. Six of the eight olfactory pores (*PorW*) in group 6 on inner surface of right hind leg of *Epilachna borealis;* also one hair (*Hr*), one hair socket (*porWHr*) and two hypodermal gland pores (*PorWGl*).

FIG. 2. Two olfactory pores (*PorAp*), five hairs (*PorWHr*) and 19 gland pores (*PorWGl*) on outer surface at proximal end of right hind leg of *Epilachna.*

FIG. 3. Five olfactory pores from tibial spine of *Epicauta marginata* (same as shown in Text-fig. 2*G*).

FIG. 4. Group 1 of olfactory pores on peduncle of elytron of *Epilachna* (same as shown in Text-fig. 1*B*).

FIG. 5. Seven of the olfactory pores in group 2 on wing of *Epilachna.*

FIG. 6. Eleven of the olfactory pores in group 3 on wing of *Epilachna.*

FIG. 7. Ten of the olfactory pores in group 4 on wing of *Epilachna.*

FIG. 8. Four of the five olfactory pores on ventral side of wing of *Epilachna.* Figs. 5 to 8 represent some of the pores as shown in Text-Fig. 1*C*.

FIG. 9. Olfactory pore from trochanter of *Uloma.*

FIG. 10. Olfactory pore from tibia of *Uloma.*

FIG. 11. Three olfactory pores from elytron of *Uloma.*

FIG. 12. Olfactory pore and sense cell from wing of *Uloma.*

FIG. 13. Olfactory pore and sense cell from trochanter of *Orthosoma* (cut slightly obliquely).

FIG. 14. Olfactory pore and about one third of sense cell (*SC*) from trochanter of *Orthosoma,* showing pit (*P*) and flange (*Fl*).

FIG. 15. Olfactory pore from tibia of *Orthosoma.*

FIG. 16. Oblique section through trochanter of *Epilachna,* showing anatomy of leg. It was cut in such a manner that no muscles are shown in the section and that the nerve (*N*) is severed in two places.

FIG. 17. Cross section through proximal end of tibia of *Epilachna,* showing anatomy of leg at this place. The gland pore (*PorGl*), hair pore (*PorHr*) and sense cells (*SC*), belonging to the tactile hairs (*THr*) were taken from two other sections, and the gland cell just beneath the gland pore was taken from the other end of this section.

FIG. 18. Four olfactory pores and a small portion of hypodermis from elytron of *Epilachna.* The material used for Figs. 17 and 18 was from an old adult beetle that had been confined in the laboratory nearly all summer.

FIG. 19. Four olfactory pores, sense cells and nerve (*N*) from wing of *Epilachna.*

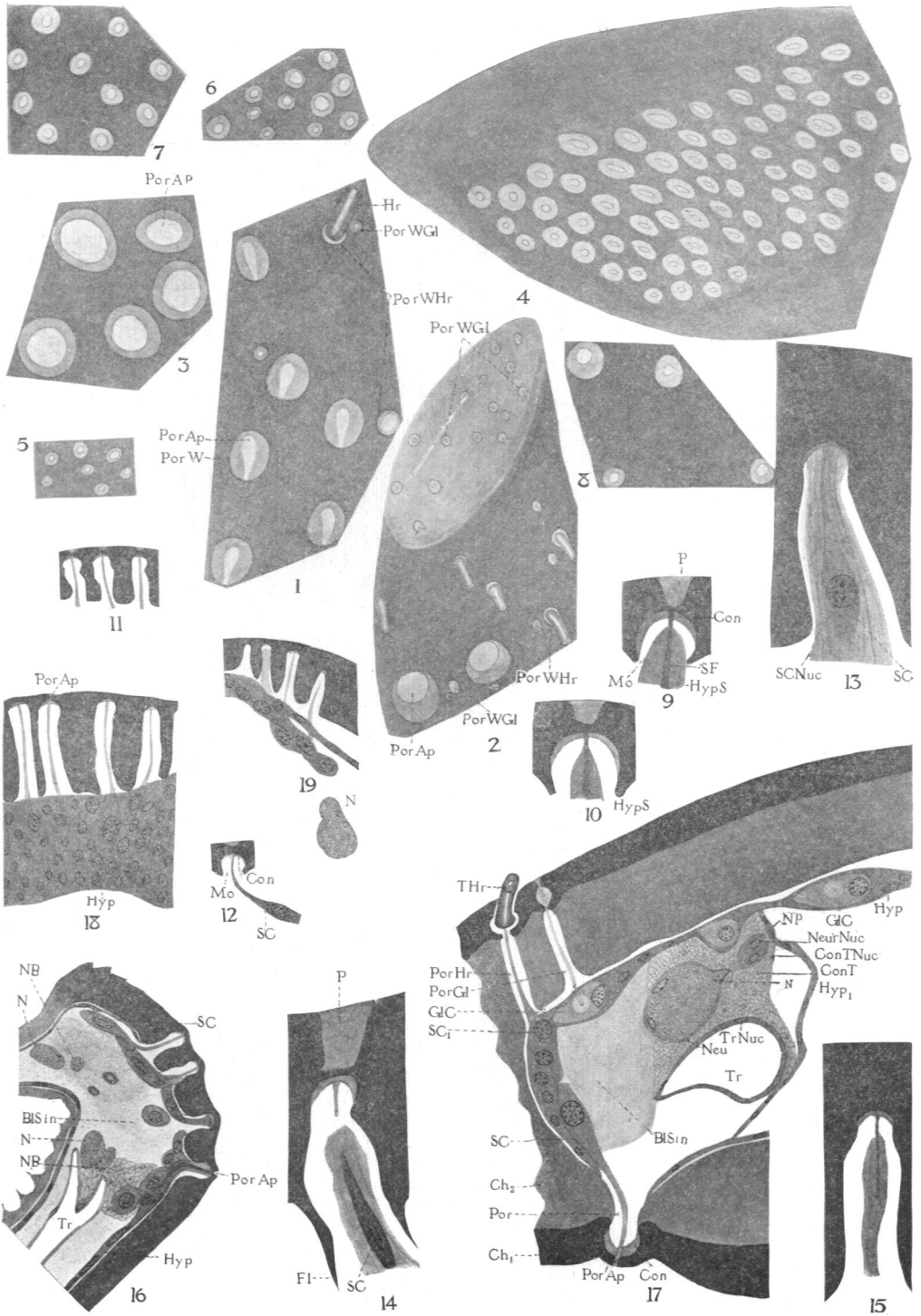

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PLATE II.

FIG. 20. Three olfactory pores from wing of *Passalus*.

FIG. 21. Six olfactory pores from elytron of *Orthosoma*.

FIG. 22. Cross section through wing of *Orthosoma*, showing anatomy of wing beneath olfactory pores.

FIG. 23. Olfactory pore from elytron of *Passalus*, showing sense cell (*SC*) connected with pore aperture (*PorAp*) and with nerve (*N*); also hypodermal cell (*HypC*) that forms the cone (*Con*).

FIG. 24. Olfactory pore from trochanter of *Epicauta marginata*.

FIG. 25. Olfactory pore from trochanter of *Epicauta pennsylvanica*.

FIG. 26. Olfactory pore from trochanter of *Leptinotarsa 10-lineata*.

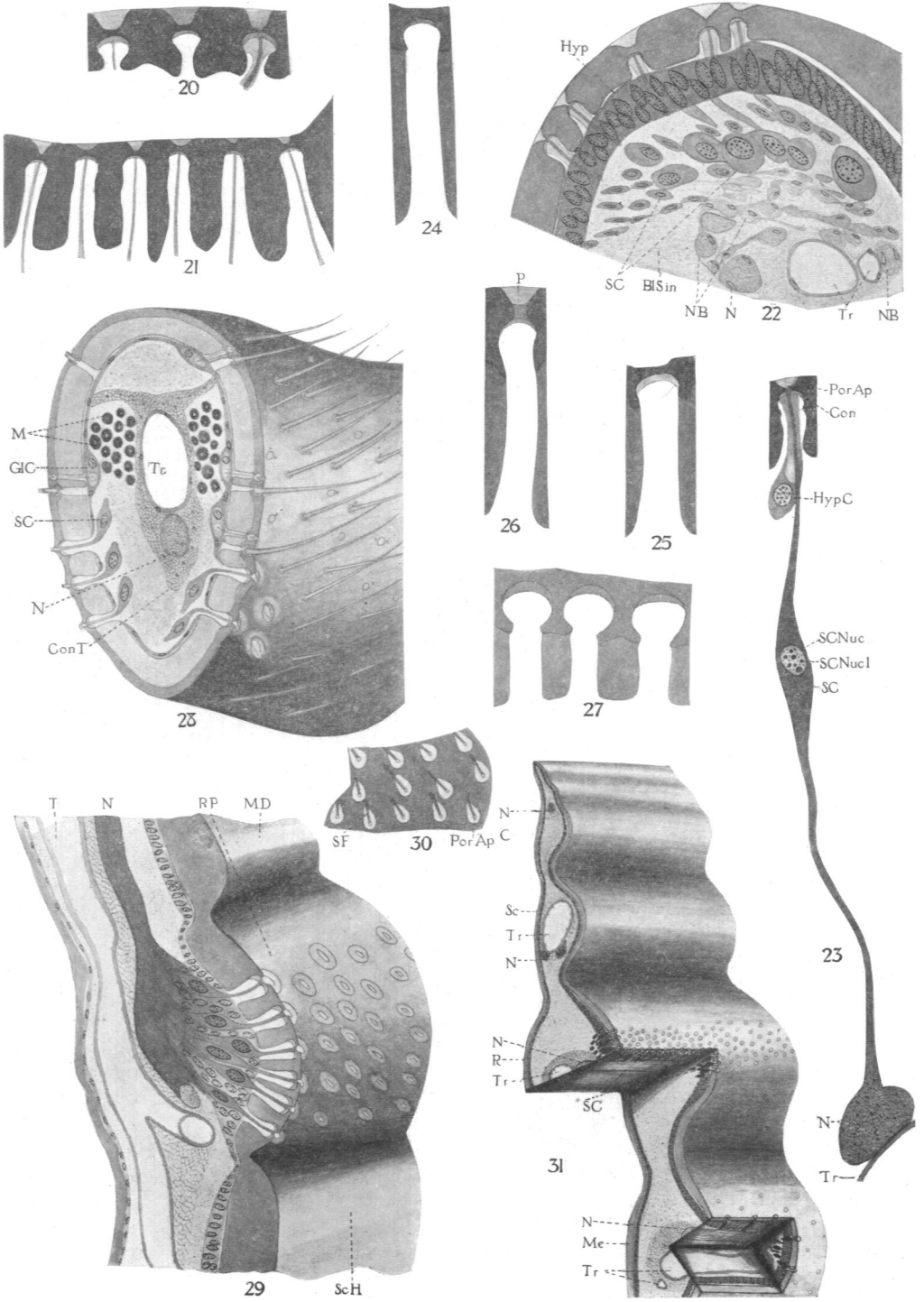
FIG. 27. Three olfactory pores from tibial spine of *Epicauta marginata*. The material used for Figs. 24 to 27 had been treated with caustic potash.

FIG. 28. Transverse-longitudinal diagram of proximal end of trochanter belonging to right hind leg of *Epilachna*, showing internal anatomy of leg and superficial view of hairs, hair sockets, gland pores and olfactory pores. The four pores at the right belong to group 6 and the three at the left belong to group 5.

FIG. 29. Oblique transverse-longitudinal diagram of portion of peduncle belonging to *Epilachna*, showing internal anatomy of radial plate (*RP*), innervation of olfactory pores and a superficial view of a few of the pores in group 1. The transverse portion of the diagram passes through the radial plate in the direction of the line marked "a" in text Fig. 1*B*.

FIG. 30. Oblique superficial view of olfactory pores on wing of *Epilachna*, showing sense fibers (*SF*) connected with pore apertures (*PorAp*).

FIG. 31. Transverse-longitudinal diagram of portion of wing belonging to *Orthosoma*, showing internal anatomy of wing, innervation of olfactory pores and a superficial view of a few of the pores on radius (*R*) and media (*Me*).



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